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BIOLOGY OF THE PARASITES AND HYPERPARASITES OF APHIDS.*

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A biological study of the parasites of aphids was begun early in the summer of 1920, after many observations during an outbreak of *Aphis gossypii* Glov. on cotton in North Carolina had demonstrated the economic importance of the parasites and predators in natural control of the pest, and had aroused great interest by their efficiency. In the course of the work, similar parasites of other aphids were encountered, and hyperparasites were found closely associated. A general investigation of these insects and their interrelations followed. Opportunity for study of the Aphid parasite Complex in Ohio was afforded in 1921, and the project was continued there until July, 1922.

In this study assistance has been given freely by many, and grateful appreciation is expressed for this help, especially to Doctor Herbert Osborn, Research Professor of Entomology, Doctor Raymond C. Osburn, Professor of Zoology and Entomology, and Doctor Clarence H. Kennedy, Assistant Professor of Zoology and Entomology of the Ohio State University, and also Doctor Zeno P. Metcalf, Professor of Zoology and Entomology of the North Carolina State College.

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HISTORICAL.

The earliest biological work with parasites of aphids was published in 1899 by Seurat. This writer studied the post-embryonic development of *Doryctes gallicus* Rhein., a Braconid parasite of certain Lepidoptera and compared the related species *Apanteles glomeratus* Linn. and *Aphidius fabarum* Marshall, the latter a parasite upon *Aphis rumicis* Linn. in France. The references to *Aphidius* are scattered through the paper, but when sought out, form a quite complete biological discussion of the development and structural modifications of the parasite itself.

Timberlake (1900) observed and described the early stages of *Praon simulans* Prov. and *Aphidius rosae* Hal., figured the first stage larva of the former, noted "inhibition" of extra larvae in cases of superparasitism, and pathological changes in what he considered the aphid fat bodies.

Ainslie (1909) discussed briefly the manner of attachment of parasitized aphids and considered that they are held to the leaves by "death grip" of the tarsal claws, or by a glutinous fluid forced through the pores of the aphid. In addition the larva afterwards rends the ventrum of the aphid and spins with its mouth a cocoon, attaching this also to the leaf through the opening, according to this author. He also recognized that aphids parasitized by *Aphelinus* are not split ventrally when attached.

Webster (1909) in a bulletin on the Spring Grain Aphis, *Toxoptera graminum* Rond. discussed control by *Lysiphlebus tritici* Ashm. (*Lysiphlebus testaceipes* Cress.) and figured the adult and egg of the parasite and also parasitized aphids. Experimental distribution of the parasite for control gave negative results. In certain parthenogenetic experiments a few females were obtained among many males in the progeny from unfertilized females.

Gahan (1911) in his systematic paper, included notes on habits, life history and hosts for each species of primary parasite of the subfamily Aphidiinae.

Webster and Phillips (1912) in a later bulletin on *Toxoptera graminum* Rond. discussed in detail the biology of *Lysiphlebus testaceipes* Cress., and of several other parasites and hyperparasites associated with these insects. This is our most comprehensive biological paper on aphid parasites.

Ainslie (1917) reported the peculiar formation of the cocoon in *Praon*, which spins underneath the skin of the parasitized aphid rather than inside the skin, as do the other parasites.

Haviland (1920) investigated the bionomics and development of *Lygocerus testaceimanus* Keiff, and *Lygocerus cameroni* Keiff., two European Proctotrypid hyperparasites of aphids through the *Aphidiinae*, and compared the stages of other Hymenoptera. The same author (1921) in an interesting paper on the post-embryonic development of three European Cynipid hyperparasites of aphids through *Aphidius* summarized briefly in a paragraph what is known about the relations between the primary parasites and aphids, and the tissue changes involved. The life history and development of the hyperparasites, *Charips* spp., were treated admirably, and comparisons were made with the development of other parasites.

Hartley (1922) treated in detail the life history and biology of *Aphelinus semiflavus* How. and certain relations between larvae of this species and larvae of Aphidius in the same aphid.

Wheeler (1923) discussed fully previous papers on the larval stages of the *Chalcidoidea*, *Proctotrypoidea*, and *Ichneumonoidea*, defined the larval types of parasitic Hymenoptera, and treated of the life histories of certain species of *Aphidiinae*, with special reference to the larval stages.

APPARATUS AND METHODS.

The insects used in this project were reared for the most part in home-made cages, each of which consisted of two six inch wooden embroidery hoops, over which was sewed a cylinder of fine-mesh cheesecloth, stretched by two thin strips of wood spacing the hoops; a square of glass covered the top and permitted observation of the plants and insects inside; the bottom of the cage was made of two glass plates which rested on the top of the pot, and which were suitably notched to fit closely around the base of the stem of the potted plant.

Aphids were grown in thse cages until quite numerous and then the adult parasites were admitted. A few aphids were removed from time to time for dissection, and at regular intervals numbers were fixed in Bouin's Picro-Formol or Brasil's Fluid for serial sectioning. Most of the insects were cut with

a microtome setting of ten micra, though some sections were made five micra thick. Delafield's Hematoxylin and Eosin, or Heidenhain's Iron-alum-hematoxylin stains were used, and the resulting slides demonstrated clearly the internal relations between larval parasites and hosts and also other points not clear from the gross dissections. The hyperparasites were handled in much the same way, or in shell vials where these were found to be suitable.

Many collections and observations were undertaken in the field, and every effort was made to understand the interrelations of the factors and species under normal outdoor conditions.

THE APHID COMPLEX.

PHYSICAL FACTORS IN NATURAL CONTROL.

From field observations both in North Carolina and in Ohio it seems that the factors concerned in natural control of aphids fall into two general classes, physical and biological. Under the first of these should be placed moisture and temperature. Moisture in the form of humidity probably exerts little direct influence on aphid abundance, but indirectly by working through certain of the biological factors to be discussed later, has considerable bearing on the problem; in the form of rain there is direct connection, as rain washes many aphids from plants and beats them into the mud where they die. Temperature is probably a more important factor. It is often observed that outbreaks of aphids seldom persist long after hot summer weather has set in. In order to throw some light upon the effect of various temperatures on aphids, a series of the cages was set up, in a chamber where the temperatures could be varied at will. It was found that *Myzus persicae* Sulz. living on dock (*Rumex* sp) and on potato plants behaved normally up to 30°C., but at 33°C. there was considerable restless activity and the winged forms flew away. At 35°C. all of the aphids were running aimlessly about and at 40°C. they began dropping from the plant; a temperature of 44°C. for ten minutes removed over half of the aphids, and a plant subjected to 49°C. for an hour was wilted, subsequently shed most of its leaves but recovered, and was thereafter free from aphids.

Comparable temperatures may be attained on sunny days in the open, close to the soil where radiation adds to the already high temperatures. In addition moderately warm periods have an indirect effect on aphid abundance, in that they encourage activity by certain of the biological agencies of control; this indirect relation is without doubt more important than the direct, as its influence is felt far ahead of the time when the extreme conditions which would bring about any direct control would be present.

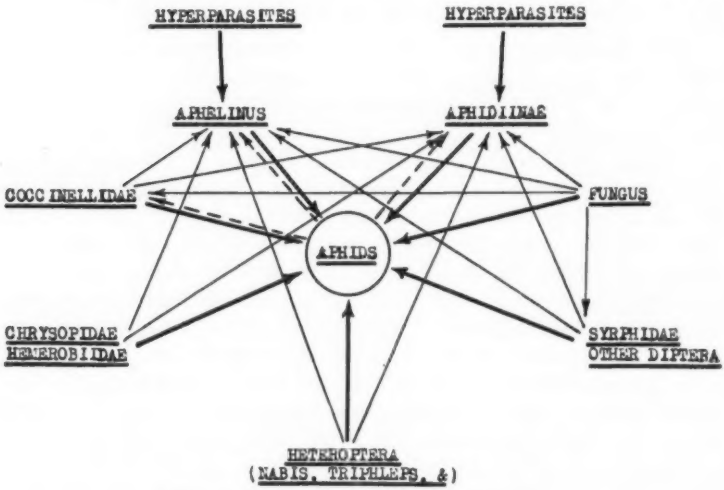
BIOLOGICAL FACTORS IN NATURAL CONTROL.

The biological factors of control include condition of food plant, fungous diseases, predatory insects, and parasitic insects. The predators count among their numbers several of the *Coccinellidae*, commonly called "Lady Bird Beetles," *Syrphidae* and other less important *Diptera*, *Chrysopidae* and *Hemerobiidae*, ordinarily spoken of as "Lace Wing Flies," a few *Heteroptera*, and a few other miscellaneous animals. The true parasites and hyperparasites with which this paper is concerned belong to the *Hymenoptera*, and are tiny wasps.

The condition of the food plant plays an important part in determining the rate of increase of aphids. Plant tissues that are succulent, sappy and growing rapidly are very favorable, as demonstrated by aphids habitually collecting on the tips of plants, and neglecting the more mature parts. Here again, wet periods play a part in favoring aphid increase, because such sappy growth is encouraged by the abundant moisture. Slow growing, well-seasoned tissues produced in dry periods are thus not as favorable to the aphids, possibly because of greater difficulty in obtaining food in proper quantity, so growth and reproduction on these are slower. For the same reason the ripening of plant tissues or the dying of the host is fatal to the aphids. This food plant relation is more or less basic for the whole Aphid Complex.

The relative efficiency of the various biological agencies of natural control varies with the time of year, and with differences in weather conditions. Early in the spring when the weather is still cool, the aphids usually get quite a start and increase in numbers very considerably. At this time of abundant precipitation, cloudiness alternating with sunshine, and moderately low temperatures, the fungous diseases of aphids

find favorable conditions and come into prominence. The Hymenopterous parasites are few in number and are not very active. Likewise, the Coccinellids, Syrphids, and Lace Wing Flies are too few to be of great economic benefit but they begin to multiply at this time. As the weather becomes warmer and more settled the fungous diseases lose first rank in control. The Lace Wings and *Diptera* seldom take prominence, but help materially the Lady Bird Beetles and the parasites. At this stage the number of aphids decreases rapidly and the



THE APHID COMPLEX BIOLOGICAL CONTROL.

Fig. 1. Heavy lines denote direct attack; light lines relation with aphid as intermediary (i. e., predator eats aphid containing larva of parasite); broken lines defense by aphid (i. e., gumming).

outbreak may subside; often after a short time the plant lice become so scarce that many of the parasites perish for lack of hosts. The predators in adult form may be able to migrate to other outbreaks of different species, as they are seldom as limited in the direction of suitable host species as are the parasites. At times there may be recurring outbreaks of the aphids of lesser extent, with subsequent reappearance and increase of parasites and predators, but usually these secondary waves are of little significance from an economic viewpoint. Often

after the primary outbreak a condition of balance will prevail for the remainder of the crop year, with a few aphids present, some parasites which are held down by hyperparasites, a few predators, and a small amount of fungus. There may be considerable competition among the insect agencies of control where aphids are few in number, and the activity of any predator or parasite may seriously restrict the opportunity of other parasites and predators, and in turn be limited by their work. Both food supply and opportunity for reproduction may be lacking when such conditions prevail. Then also, there may be direct destruction of beneficial insects by other beneficial insects, as where the young stages of the parasites inside of the bodies of aphids are eaten when predators devour such plant lice. The complex relations of the many factors concerned in biological control are shown graphically in Text Figure 1.

THE PARASITES OF APHIDS.

The Hymenoptera associated in the Aphid Complex are divided, because of their habits, into two groups: those parasitizing aphids directly, alluded to as the "Primary Parasites" or simply "Parasites," and the forms attacking stages of the primary parasites in aphids, spoken of as the "Secondary Parasites" or "Hyperparasites." To complicate relations still more, there is good evidence that certain of the hyperparasites at times attack other hyperparasites or even young stages of their own species. This is rarely accomplished in the Aphid Complex under natural conditions and probably has little economic bearing but nevertheless it is a relation of great interest. It is evident that the Primary Parasites are vastly beneficial in an economic sense, since they limit the reproduction and bring about the destruction of insect pests that are most troublesome. As the Hyperparasites in turn limit the activities of the Parasites, they are to be considered as harmful insects.

The Primary Parasites are assigned by systematic workers on the Hymenoptera to two diverse groups: (1) *Ichneumonoidea*, Family *Braconidae*, Subfamily *Aphidiinae;* (2) *Chalcidoidea*, Family *Aphelinidae*. There are many species of the first group, the *Aphidiinae*, found wide-spread thruout this coun-

try and the world; they are most active and prolific and are by far the most important of aphid parasites. When compared with their work, that accomplished by the second group, the *Aphelinidae*, is insignificant.

The Hyperparasites are scattered even more widely by systematists than are the Parasites. There are forms assigned to the *Chalcidoidea*, Families *Entedontidae* and *Pteromalidae;* some *Cynipoidea* belonging to the Family *Figitidae*, and a species of *Serphoidea* (*Proctotrypoidea*), family *Ceraphronidae*.

In reporting the results of this study a general discussion of the insects belonging to each group will be given; in the case of the *Aphidiinae* the general discussion of habits, development, and economic importance is based upon a thorough study of *Diaeretus rapae* Curt., *Lysiphlebus testaceipes* Cress., *Aphidius nigripes* Ashm., *Aphidius polygonaphis* Fitch, and *Praon simulans* Prov., and the data obtained thereby has been checked up as far as possible with the other species mentioned in the body of the paper. The other groups of parasites have been treated in a comparative manner also where this was possible.

THE APHIDIINAE.

HABITS OF THE ADULTS.

The life activities of the adult parasites are very interesting. Just after emerging from its cocoon in the dried aphid skin the parasite is soft and rather wet, but it requires only about five minutes for the wings to expand and dry and the body to become compact. During this period the movements of the insect are rather uncertain and poorly coordinated but it is surprising how soon the adult becomes fully mature. The adults are most active on bright sunshiny days when the temperature is between 20°C. and 30°C. At lower temperatures the parasites remain on the under sides of the leaves, more or less inactive. They are extremely sensitive to variations of humidity; excessive dryness will kill both parasites and aphids quickly, as has been demonstrated many times in cages and breeding vials. There is little activity among them on dark or rainy days or at night when they rest quietly on the lower leaf surfaces.

In the field the problem of obtaining food presents no difficulty to these insects. They have only to lick up the honey dew excreted by the aphids. In this respect they are messmates of a host of other Hymenoptera such as ants and many kinds of wasps. However the parasites do not "milk" the aphids and eat the honey dew direct from them, as some insects are reputed to do, but lap it up from the top of lower leaves which often become varnished over with the material dropped from above. In the cages and vials used in the breeding work, a mixture of clover honey and water was used for feeding, and all of the parasites lapped this up greedily until gorged.

Wintering: The method of wintering of the Aphidiinae parasites depends more upon the resistance of the plants on which the aphids multiply than upon the severity of the cold. The adult parasites are able to withstand temperatures that are surprisingly low. In experiments, adult *Diaeretus rapae* have been kept out-doors with no protection except a small glass vial in which they were confined, for periods of over thirty days, alive and well, but of course inactive, with temperatures ranging from -12°C. to 10°C. When these parasites were brought indoors, within fifteen minutes they were as active as in the summer. It was found to be impossible to carry any of these through the entire winter, however, as they became emaciated after about a month and died from starvation. The pupae of the parasites are even more resistant and serve to bridge over the winter, at least in the species mentioned. For example, on February 4, 1922, examination was made of the plants in a cabbage patch at Columbus, Ohio. The field had been cut over several times in the fall and most of the heads sold, but here and there were left poor plants that had grown somewhat, but at the time mentioned were badly injured by cold and half covered with snow and sleet. A half dozen plants revealed one live and thirty-two dead parasitized *Aphis pseudobrassicae*, which were brought into the laboratory. In five days six perfectly healthy *Diaeretus rapae* had emerged from those parasitized. They had been subjected to all sorts of weather in the winter, and it is surprising that so many were alive. Cold merely seemed to slow up their development. Undoubtedly the natural refrigeration of the pupae in winter serves to tide over this species until spring.

Aphids, if they are able to obtain food, can reproduce in sheltered locations even in very cold weather, but if the hosts die at first frost as do tomatoes or melons, the aphids upon them succumb to starvation and the parasites cannot survive; or if the plants ripen in hot weather as do the grains, the aphids may be destroyed just as thoroly by lack of food and the parasites find no hosts upon which to work. In such cases the parasites do not remain in the locality all year around. On the other hand, in places where growing conditions are more favorable and where plants do not die from frost or ripen too soon, the parasites are able to continue their reproduction and migrate in waves when conditions are suitable again. This was the case with *Lysiphlebus testaceipes* in 1921. In the fall the first frosts kill the melon, squash, or other host plants long before settled cold weather, and the remaining pupae transform into adults which finally perish. No viable pupae remain to be refrigerated over the winter as with *Diaeretus rapae* because settled cold weather does not follow soon enough after the first killing frost. In regions of the South, such as Florida or Texas, melons or cotton or other host plants may not be killed out, and may support the aphids and the parasites all winter. Then in the spring there is a wave of aphid migration northward, and a subsequent wave of parasites. For example, in 1921 *Lysiphlebus* reached Raleigh, N. C., on June 10, but did not appear at Columbus, Ohio, until September 6; *Aphidius nigripes* reached Raleigh on May 7 and disappeared on June 1, but reached Columbus early in June and disappeared by July 15.

There is one other possibility of wintering in the North. Greenhouses often become infested with aphids and it is possible that parasites would survive under the protection of these structures. Such a possibility is a remote one, although there is good evidence that in the winter of 1921–1922 *Aphidius phorodontis* came through in this manner in a conservatory at the Ohio State University upon aphids of tomato and other plants.

Mating: The females and males are ready for mating as soon as they have dried out after emergence. To sense the presence of the female the male has to be within a short distance of her, seven to ten millimeters being the limit. The female when approached usually tries to run away and the

male pursues and climbs on the back of the female, which seems to have a quieting effect upon the latter. Then the male crawls backward, curves the end of the abdomen over and under the tip of the abdomen of the female, and copulation ensues. This lasts from a few seconds to a minute and is usually terminated by the female breaking away and crawling off. Often, if many males are present, extra males will mount a mating pair and endeavor to copulate also; sometimes these will persist in their efforts to mate with the successful male long after the female has left. Several instances have been observed where a female was carrying about three males, one above the other.

Oviposition: Oviposition may occur with or without previous mating and fertilization of the female, and the response of a female to the presence of a suitable aphid is equally vigorous, whether virgin or mated. The act of oviposition has been described and illustrated by Webster (1909) and many other authors have copied or adapted his illustrations. Briefly, the process is as follows: The parasite comes up to an aphid, arches her antennae above it and taps it a time or two with each antenna; then if apparently satisfied, she curves her abdomen under her head and thorax, between the braced legs, and still facing the aphid, elongates the abdomen until the tip comes in contact with the body of the aphid; then there is a quick forceful stab and oviposition has been accomplished. Usually only one egg is placed in an aphid but where there are few aphids, as in certain of the oviposition experiments carried on in glass vials, or a superabundance of parasites, an aphid may be "stung" several times. The egg may be placed in any part of the body of the victim, even in the head. The aphids seem to suffer little or no inconvenience from the oviposition; a drop of exudate appears at the point of puncture but this soon hardens and no observable derangement is produced, except where the aphid has been oviposited in many times, when a slight sluggishness is manifested. For oviposition the *Aphidius* parasite ordinarily selects a half grown aphid, but when conditions are such that egg laying is vigorous, as on quiet, hot, sunny days, the parasite will attack aphids of any stage and will lay egg after egg in one individual if the supply of aphids is limited.

The process of oviposition is not entirely without danger to the parasite. If any part of the insect comes into contact with the cornicles of the aphid, it becomes "gummed" with the gluey secretion produced by the plant louse. This secretion hardens on the parasite and renders it temporarily "*hors-de-combat.*" The parasite will then spend half an hour or so scraping at the affected parts in an effort to remove the substance. Such "gumming" often produces permanent impairment of the functions of the tiny wasp, and may hasten death by several days.

THE EGG.

The mature ovarian eggs of these *Aphidiinae* are elongate oval structures with one or both of the ends tapering more or less into blunt projections. When completely developed in the ovary the measurements average, with *Diaeretus rapae*, 50 by 20 micra. In oviposition these are placed by the parasite in the adipose tissue of the aphid, usually with part of the egg projecting a short distance into the body cavity. Almost at once the egg absorbs liquid from the aphid by osmosis, and increases enormously in size, becoming lemon shape and as large as 130 by 60 micra. Plate VII includes drawings of ovarian eggs, Fig. 1, and eggs after oviposition and absorption of fluid from the aphid, Fig. 2.

EMBRYOLOGY.

The early stages in the development of species of *Aphidiinae* are exceedingly difficult to follow, for several reasons: the embryos are very small, their cells are densely packed together, the tissues are poorly defined, there is little or no yolk material, and it seems that many of the usual processes of embryonic development are either modified greatly or eliminated entirely, being bridged over by these parasites.

The youngest embryo obtained was a stage of *Diaeretus rapae* of approximately ten hours development. This lay in the adipose tissue of the aphid completely enveloped, and measured 30 by 50 micra. The chorion had disappeared except for a small sector originally around one of the projecting ends of the egg. The embryo itself contained many nuclei, arranged radially from the periphery toward the center, two or three deep. Between adjacent radii were noticeable divis-

ion lines reaching nearly to the clear center area. The most notable thing about this embryo is the light thrown by it upon the formation of the serosa, the only membrane formed by the embryo in these species. This embryo is shown in section as Figure 3.

The Serosa: At two or more points on the periphery of the middle section on the slide can be seen round cells, exactly like the cells in the embryo, but pushed outside of the outer layer. Adjacent to these are similar external cells partly flattened out and joining each other with their thin cytoplasm to form the membrane. Cells in all stages of transformation from the spherical ones just extruded to the typical flattened serosal elements are shown by this section. Thus the serosa is formed by delamination or extrusion of cells from the ectoderm of the embryo early in embryonic development. These flatten, grow both in amount of cytoplasm and in nuclear material, become polyhedral finally, and spindle-shaped in cross-section. The nuclei become oval in outline, considerably flattened, many times the size of the embryonic nuclei, and more open in construction as the chromatin masses are well separated. The cytoplasm is finely and evenly granular and is inclined to take the hematoxylin stain rather than the eosin. Stages in the formation and the later development of the serosa are shown in Figures 15, 16, 17 and 18.

Embryos of 24 to 48 Hours: Because of the methods of taking aphids for sectioning at regular daily intervals, the next stage in development, the embryo of twenty-four hours, appears many times in the slides of all species of *Aphidiinae* studied. In this there is considerable increase in size, in number of cells, and the serosa is completely formed and has grown to the point of fitting rather loosely around the embryo. The radial divisions between groups of cells are quite distinct, and the central space which is the segmentation cavity still persists. This embryo has a wonderful similarity to the "morula" stage so much stressed in some courses in Vertebrate Embryology. It shows little advance over the earlier embryo. Figure 4 is a section of an embryo of this stage, in its membrane

During the second day the embryo elongates considerably in the future anterior-posterior direction, by cell multiplication; the segmentation cavity elongates likewise, but keeps the same diameter. The head region grows faster than the other

parts and from its greater diameter can be recognized. However, no tissue differentiation can be made out in any of these embryos, except that the cells of the outer layer appear very slightly smaller than those in the interior. In this period the rapid growth is seldom uniform throughout the body of the larva, and this unequal development brings about peculiar twistings so that the embryos appear for all the world as if they were "writhing in agony." The serosa is still present, having enlarged somewhat in the interval. This stage and the twenty-four hour and many intermediates were found in slides of *Diaeretus rapae*, *Praon simulans*, *Aphidius nigripes*, and *Aphidius polygonaphis*. It is delineated in Figure 5. A few hours more and the young larva untwists and takes a position in its membrane so that the head and posterior ends are approximated, and the body is looped in a quite regular curve; this is accomplished by an equalization of growth, not by the action of muscles. From this point onward development seems more rapid.

Embryos of 48 to 72 Hours: During the third day the size of the head increases enormously, external segmentation of the body occurs, and tissue differentiation is initiated. The serosa has become quite roomy and the abdomen of the embryo in growing uncoils more and more. In the early stages of the third day the nervous system mesenteron, proctodeum, the two Malpighian tubules, and the two salivary glands can be recognized as distinct structures, having become less densely packed together and separated by spaces connecting with the segmentation cavity. At this time the cells of all these organs are alike as to size, appearance, and staining capacity, and have not reached the point of specialization, appearing like those in the earliest embryo found. However, this specialization occurs by the end of the third day and then and thereafter the organs change little except in size, up to the last molt. Figures 6 and 7 represent embryos of the third day, reconstructed from serial sections.

Hatching: Some time during the third day the larva frees itself from the serosa and comes to lie in the body cavity of the aphid. It is possible that the membrane is torn asunder by the larva growing and becoming straighter, but it is more probable that the larva, beginning at the time of tissue differentiation, secretes a cytolytic enzyme that aids in breaking up

the serosa. Possibly there is a combination of these two processes. Certain phenomena to be discussed later lend strength to the enzyme hypothesis. It is a fact that hatching is not the result of violent muscular movements, since the muscles are just starting to develop, and furthermore the aphid tissues are never disturbed in the least, as would occur with activity of the larva of such a nature.

THE FIRST LARVAL STAGE

The larva of the first stage (Figure 8) is elongate and uncurved, or but slightly curved as it grows older, and is composed of a large head and thirteen body segments that taper gradually to the posterior end, which ends in an elongate "tail" in all species of *Aphidiinae* studied. In *Praon* this tail is double, or rather bifid. It has been suggested that this structure functions in aiding the larva to move about inside the aphid, or to prod about into the tissues of the latter to hasten disintegration and release food for the parasite. Both views are erroneous, for up to the time of the last larval instar there is no indication either in the larva itself or in the tissues of the aphid, that there is any voluutary movement of the larva, and there is certainly no mechanical change in the aphid tissues to substantiate the prodding theory. In the first larval stage, as in the embryo, absorption, respiration, and excretion are evidently carried on by the cells of the ectoderm of the parasite, by the process of osmosis. The food is carried to the larva in the blood of the aphid, and the oxygen likewise, and wastes are carried away by the same medium. The larval parasite develops in precisely the same manner as do the embryos of the aphid, and so there is little need for moving or foraging for its food.

The Fate of the Serosa: At the time of hatching the serosa is broken up into pieces, sometimes as many as thirty being left in the body cavity of the aphid, each piece varying in size and in number of nuclei. The fragments of the serosa round off, and begin a strange process of vacuolization and growth, assuming forms as in Figures 15, 16, 17, and 18. The nuclei enlarge also, break down, fuse and finally result in a shrunken mass of chromatin. The cytoplasm becomes more and more vacuolated as the parasite larva develops, and the size of the degenerated mass increases steadily until the larva is ready

for the last molt. What happens then to these degenerate masses will be discussed later. They have been observed by Timberlake (1900) and many others but their origin has been universally misunderstood, as they have been referred to the aphid fat bodies for their origin, and the process by which they arise has been termed "pathological changes in the fat body." The fat body of the aphid is modified by parasitism but in an entirely different manner, as will be explained in the following pages.

Inhibition of Younger Larvae: It has been noted by the author mentioned above and by others that only one adult parasite ever emerges from a parasitized aphid, even tho sevdral eggs were laid therein at the same time or at different times. To account for this the theory was advanced that in some manner all but one of the larvae are inhibited or suppressed by the successful parasite. This much is true, as the serial sections of aphids that have been superparasitized in this way substantiate these facts, and in addition suggest a probable manner of accomplishment. Shortly after hatching, the first stage larva is seen to be growing vigorously and rapidly. But younger larvae in the same aphid soon lag behind with their growth checked; their bodies shrink, the contours and organs lose their definiteness, and the cytoplasm of the cells seems to be abstracted, leaving the nuclei very prominent by contrast, and closely packed together. One glance at such a larva is enough to satisfy one that extensive destructive changes have taken place, and that the process of inhibition is a bio-chemical rather than a mechanical one, for never are there any evidences of mechanical injury such as would be present if the older larva had eaten parts of the younger, or otherwise molested it. Suppressed and normal larvae of the type discussed are represented in Figures 23 and 24.

Effect of Parasitism upon Adipose Tissue: Almost as soon as the parasite larva breaks out of the serosa the neighboring adipose tissue becomes affected. The cell boundaries become more pronounced, the cytoplasm more and more vacuolated, the nucleus shrinks and instead of being rounded takes on a very irregular outline. It would seem that the substance of the adipose tissue is sapped by the developing parasite and the cells are left in a starving condition. As the larval

parasite develops, the sphere of affected adipose cells correspondingly increases, until all of the tissue in the aphid is involved. From all indications in the serial sections the associated oenocyte cells are not affected in the least. A similar change in the adipose cells may be observed in old aphids that are producing a number of large embryos, but seldom is as much effect seen as is invariably present with parasitization.

The manner in which this destructive effect is produced is not entirely clear. It is certain that the young larva does not directly eat the tissue, since the serial sections show no evidence of the process, and the mesenteron of the parasite is absolutely empty of solid food and even of yolk, which we do not find in the embryo at any stage. The food is absorbed by the larva from the body fluids of the aphid, in a manner entirely comparable to that by which the aphid embryos get their own food. Seemingly the larval parasite has need of much more food for its growth than has a group of aphid embryos, if we judge on the basis of the extent of tissue involved. Undoubtedly adipose cells function in insects as storehouses of food, and serve to keep the available supply of this in the body fluids at a condition of balance. That is, if an insect must exert itself extraordinarily in travelling, or needs much food for transformation or the process of reproduction, that is supplied by the blood, and the blood is replenished from the adipose cells. When the period of stress is over and the insect feeds again, the soluble food in the blood becomes more concentrated and the adipose tissue builds up again from this excess. It is very evident that a parasite larva utilizes a vast supply of food at the expense of the aphid, to the point where all the adipose cells are starved. Figures 19 and 20 illustrate the effect of parasitism upon adipose tissue of aphids.

Effect of Parasitism upon Reproductive Tissues: The most remarkable changes in the aphid due to parasitism are to be seen in the tissues of the reproductive system. The embryos are especially modified; in whatever stage they may be at the time of hatching of the parasite they are arrested in their development, and as the larva of the parasite grows, large vacuoles and spaces appear in the embryonic tissues, the cytoplasm of the individual cells becomes thin as evidenced by difference in staining capacity, and the embryos finally degenerate into vacuolated masses. Strangely enough the

pseudova before the initiation of division of cells to form embryos are not affected until the young and old embryos have been rendered entirely unrecognizable. Camera lucida drawings of aphid embryos in two stages of degeneration from parasitization are shown in Figures 21 and 22.

Like the pseudova, the ova which develop into winter eggs in some aphids are arrested in development by the larval parasite, but show only slight change in structure. In some cases a thinning out of the yolk material is to be seen, but nothing more. From an economic point of view, the fact that parasitization brings about a cessation of reproduction of the aphid on the third day or thereabouts is an important consideration, since aphids may produce several young per day, and these are suppressed a week before the aphid itself succumbs to the parasite. In reckoning the economic value of these parasites in comparison with other control factors it would be very unfair not to give this point due weight.

Effect of Parasitism on Other Aphid Tissues: As far as could be observed parasitism causes no change in the muscular or nervous tissues. The digestive system in some cases seems to escape likewise, but at times there is to be seen in the serial sections a little of the same dissolving effect. It is of great advantage to the parasite larva that the nervous, muscular, and digestive systems of the aphid should remain intact until late in the process of parasitization. The last system furnishes food during all this time for the parasite, and the untouched muscles and nerves of the aphid mean that the latter stays on the plant and feeds, and does not fall off, starve, dry up, and die, leaving the unfortunate half developed larva inside to perish also.

MODE OF PRODUCTION OF ABNORMALITIES.

The means by which the larval parasite is able to produce such profound effects upon the cells of its own serosa, the younger larvae of its own species in the same aphid, the adipose tissue of the aphid, the embryos and pseudova of the latter, is worthy of careful consideration. It is clear that none of these can be mechanical effects, because the "tail" of the larva does not lacerate the tissue and indeed the larva does not move at all in the aphid in this stage. There is more involved than a mere starvation, since certain of the tissues concerned

show growth and peculiar hypertrophy. The theory of the writer is that the first larva arriving at the hatching stage molts and then begins to secrete a cytolytic or digestive enzyme, which frees it from its serosa, brings about the degeneration of the younger larvae, abstracts food from the aphid fat body, saps the developing aphid embryos and pseudova, in short, brings about the many cytolytic changes observed in connection with parasitism. The enzyme seems especially active upon growing and unstable tissues and affects only slightly the more stable parts such as digestive system, the nerves and the muscles of the aphid. The larva secreting this enzyme probably protects its own tissues by elaborating anti-ferments in the cells, as the cells of the vertebrate stomach are supposed to produce against the digestive ferments, which would otherwise digest the stomach itself.

THE SECOND STAGE LARVA

It is extremely difficult with species of *Aphidiinae* to determine with accuracy just when and where the molts occur in the process of development, since the stages are all internal and none are heavily chitinized, so the delicate shed exuviae are almost impossible to see even in the stained slides. For this reason differences in size, posture, development, body shape and internal organization have all been resorted to in order to determine with some accuracy the various instars. The second stage larva, Figure 9, is found ordinarily during the fifth day of development, although this time varies considerably for the different species, and even for larvae of the same species, for larvae in large aphids with an abundance of food develop more quickly than those under more adverse circumstances, and variations in temperature also seem to influence the rate of growth a great deal. This second stage larva is distinguished from the first stage by its larger size and by the greater curvature assumed by the body; also the anal process or "tail" is larger but more blunt than in the preceding larva. Nutrition and respiration, as in the previous stages, are entirely cutaneous, as there is no connection yet between the mesenteron and the outside, and the tracheæ have not reached a point of development that would permit functioning. Part of the excretion may be accomplished by the Malpighian tubules, as these seem mechanically capable of discharging their function at this stage.

THE THIRD STAGE LARVA

The distinguishing features of the third larval stage, Figure 10, which is found in some species about the sixth day, is the inflation of the abdomen by growth, so that the tail is no longer prominent. Internally the mesenteron has become much more capacious, but remains empty and shut off from the exterior. The salivary glands by their intense staining reactions reveal great activity, and two mandibles appear at the mouth, from which the stomodeum leads half way to the anterior wall of the mesenteron. Nutrition, respiration and excretion are still the same. The vacuolated masses derived from the serosa are now very large, surround the larva, and probably act as cushions in giving it mechanical support and protection. Until the end of this stage the aphid continues its vital processes in a more or less normal or subnormal fashion. Its reproduction was suppressed on the third day, and peculiar progressive internal changes started then, but in spite of this it appears to be normal enough that one could not definitely say from external evidences, whether or not it had been parasitized.

THE FOURTH STAGE LARVA

At a certain point in the development of the nearly grown larva there is a molt and a complete change of relations. The passive larva, fed by absorption and obtaining oxygen in the same way, becomes very active and commences to eat out the internal organs of the aphid with its well developed mandibles. When this begins the aphid moves no more, but grips the leaf in a last effort. The embryos or ova disappear first, then the digestive tract, the adipose tissue, the degenerated masses derived from the serosa of the parasite and the aphid embryos, the muscles of the thorax and abdomen, and the nervous system. The appetite of the larva, and its frantic efforts to satisfy this, are striking to behold through the semi-transparent aphid skin. In half a day or a day the aphid skin is empty, except for the larva and the parts in legs and head that could not be reached. During the eating-out process the plant louse turns a darker pearly color that is quite characteristic. Before the aphid skin begins to dry the larva chews away the entire area of ventral skin that is applied to the leaf surface.

In the larva other changes take place. The mesenteron previously small and empty, expands to twice and thrice its former diameter to accommodate the ingested food. The spiracles begin to function and the salivary glands become even more prominent and active, as evidenced by their more intense staining reactions. Before the aphid skin dries, the larva begins spinning its delicate silken cocoon inside this from the salivary glands, squirming and writhing the while with the result that the threads are quite evenly distributed. The skin of this stage is closely set with small tubercles, which probably grip the aphid skin during the squirming and thus assist the larva in moving about inside the shell. The silk adheres to the inside of the aphid skin and to the leaf in the spot where the skin has been devoured, and hardens quickly. Strand after strand is added in a hit-and-miss fashion until there is a uniform opaque layer on all sides. The space ventrally, where the silk is in contact with the leaf surface, is an added anchorage for the parasitized aphid and forms a final means of attachment, as the legs of the aphid soon dry, warp, and lose their hold. The fourth stage larva, Figure 11, now rests a day as if exhausted by its great activity, voids the undigested remains of its gluttonous meal as a meconium, Figure 12, and starts transforming into a pupa.

THE PUPA

The pupa, Figure 13, upon attaining complete development, transforms into the adult, Figure 14, inside of the cocoon. With the sharp mandibles the latter chews a neat circular hole thru the aphid skin, just large enough for it to crawl thru into the open. Sometimes the circle cut out is left attached by a small hinge and looks like a lid or cover but more often this is lost, either being cut loose by the parasite or being broken off in the process of emergence. There is left behind the dried aphid skin, and inside of this a string of about six to a dozen small black objects, very much like a miniature string of short sausage links, which are the remains of the dried meconium, Figure 12.

THE APHELINIDAE.

The majority of the species of the genus *Aphelinus* are parasitic upon scale insects, and only a few attack aphids. In the course of these investigations the blackened aphid-skins containing pupae of *Aphelinus semiflavus* Howard were encountered twice during 1921 at Columbus, Ohio, both times a single example in *Aphis pseudobrassicae* Davis upon kohl-rabi. No other species of the Family were obtained. This poor showing of *Aphelinus* is not difficult to explain: first, this parasite is not as vigorous in oviposition as the more active *Aphidiinae*, which destroy many more aphids; second, in respect to distribution, *Aphelinus* is a poor traveller, being disinclined or unable to use its wings for flight; the individual seldom moves quickly, except to hop like a flea when disturbed; lastly, a female will seldom attempt to oviposit in a moving aphid, or an old aphid, seemingly preferring the younger and weaker stages, and thus the reproduction of the old stem mothers is not curtailed as is often the case with those attacked by the Aphidiine parasites.

The biology and life history of *Aphelinus semiflavus* Howard have been studied recently by Hartley (1922) in considerable detail. This writer states that the winter is passed in the pupal stage, and the adults after emerging have been kept alive as long as 39 days, producing as many as fifteen eggs a day during part of this time, though the average is much less. Males are rare and parthenogenesis is the rule, females being produced without fertilization. Feeding is usually accomplished by the female sucking aphid juices thru ovipositor puncture holes. The eggs measure .21 by .05 mm., are oval and taper at each end. They hatch in three days and the larvae complete development in from five to nine days, feeding internally with their mandibles this while on the organs of the aphid. The author adds to the list of host aphids *Macrosiphum pisi* Kalt., *M. granarium* Kirby, *M. sanborni* Gill., and *Anuraphis viburnicola* Gill. Larvae of *Aphelinus* in aphids with larvae of *Aphidius* are arrested in growth and disintegrate; as reasons for this cannibalism, starvation, toxic substances, and lack of oxygen are suggested as possible causes and discussed. *Asaphes americana* Gir. and *Alloxysta* sp.

are mentioned as hyperparasites. Excellent figures of the adult in the act of oviposition, and of developmental stages are included.

The work done by the present writer in connection with *Aphelinus semiflavus* substantiates the observations of Hartley in every essential respect, and in addition throws light upon several minor points not brought out by that worker. Serial sections of aphids infested with larvae of both *Aphelinus* and *Aphidius* show that the former are arrested in development and caused to disintegrate not by starvation or lack of oxygen, which as Hartley says would surely affect all the larvae and not just the *Aphelinus*, and would not explain the fact that when two *Aphidius* larvae and one *Aphelinus* larva are in the same aphid, only one *Aphidius* grows and the other and the *Aphelinus* are suppressed; nor by cannibalism since the *Aphidius* larva does not use its mandibles until long after the effect has been produced, and the larvae are always uneaten in the serial sections; and not by toxic substances in the sense of poisons or the like, since such agents would hardly be innocuous to one *Aphidius* larva and destroy the other and the *Aphelinus*. The true state of affairs as revealed by the serial sections is this: one *Aphidius*, the older, appears vigorous and like all rapidly growing tissues takes a deep stain. The disintegrating larvae of both species show their cytoplasm very weakly stained as if nearly all of that substance had been abstracted. The nuclei are dark and show no change but appear by contrast very clear and distinct. As explained previously the older *Aphidius* larva upon reaching a certain stage in development evidently starts secreting a cytolytic enzyme which affects the tissues of the *Aphelinus* larva, younger *Aphidius* larvae, and aphid tissues. Thus the *Aphidius* eliminates competition by secreting a cytolysin which abstracts essential materials which it probably uses for its own growth, and not by means of a toxic substance in the sense of a poison.

Sections of aphids with *Aphelinus* larvae "in situ" are delineated as Figures 33 and 34.

THE HYPERPARASITES.

Lygocerus niger Howard.

Lygocerus niger Howard was secured in the field from parasitized *Macrosiphum rudbeckiae* and *Macrosiphum ambrosiae* on ragweed, *Ambrosia trifida* Linn. and Jerusalem artichoke, *Helianthus tuberosus* Linn. in September 1921, when it was found to be associated with *Aphidius polygonaphis*. Laboratory experiments with *M. rudbeckiae* of various ages, unparasitized and parasitized by *Aphidius polygonaphis*, showed that *Lygocerus* oviposits only in those parasitized lice from which the internal organs have been entirely eaten out; that is, *Lygocerus* is a hyperparasite and attacks *Aphidius polygonaphis* in the last larval and early pupal stages. Laboratory experiments indicate that *Lygocerus* will also parasitize similar stages of *Diaeretus rapae* and *Aphidius phorodontis*. In these three hosts seven generations have been reared in the laboratory up to January, 1922, from the original stock.

An interesting relation has come to light in connection with the laboratory breeding work. Several times upon dissecting stages of *Lygocerus* out of the aphid skins young pupae and full grown larvae of the species have been found which bore eggs of *Lygocerus* upon them. These eggs develop normally into adults, certainly a peculiar circumstance for the species to parasitize its own younger blood brothers and sisters. Thus *Lygocerus* is not only a Secondary Parasite but may be a Tertiary Parasite under these conditions. It is entirely possible that the Tertiary Parasite in turn could be parasitized in the same manner, and so on until so much of the original amount of food was used up in the process that the last parasite would not have left enough to complete development.

The adult *Lygocerus*, Figure 32, is an active nervous parasite, differing greatly in appearance from the other forms encountered. In ovipositing the female searches over the aphid community with her antennae, which are never still, until one of the rounded inflated aphid skins containing an *Aphidius* larva or pupa is found. Over and over this she crawls, investigating it thoroly with quick, continuous, alternating tapping of the two antennae. If apparently satisfactory, the female mounts the parasitized aphid, bends the tip of the

abdomen to touch it, and starts drilling in with the ovipositor. When fully exserted the ovipositor is unsheathed until its base at the middle ventral part of the abdomen is exposed, and the entire length of the apparatus is pushed into the aphid skin. Differing with *Aphidius* again, where oviposition is but the act of a second, *Lygocerus* consumes several minutes, seemingly exercising great deliberation and care in the process.

The egg of *Lygocerus niger*, Figures 25 and 26, when newly laid measures 0.25 by 0.10 mm. In one plane it is oval, with one end tapering more than the other; in the other it appears curved a little, being slightly concave at the place where it is applied to the body of its host. The surface is finely striated in the direction of the long axis and at each end there is a small teat-like projection. The egg is ordinarily attached somewhere near the middle of the body of its victim, well down toward the ventral side, in the curvature produced by the cramping together of the head and posterior end of the larva or pupa in the aphid skin. Usually only one egg is laid in a parasitized aphid, but two have been observed several times. In the latter case the extra egg is not placed adjacent to the one first laid but apart from it at some other point on the body of the host, several segments away. In the course of development one larva of the two gets the upper hand and consumes the other, so that only one adult issues. After the normal oviposition, incubation of the egg takes from a few hours less than a day to a full day. Some hours before hatching takes place the digestive tract and other organs of the developing larva can be seen thru the chorion.

In hatching the larva eats its way out through one end of the egg, and the chorion is left in place, much wrinkled. The first stage larva, Figure 27, is the same length as the egg, and tapers posteriorly; the mandibles are on the ventral side of the head and are slender, small, thorn-like structures, very sharply pointed. When exposed to the light by dissection, much activity is displayed by this larva, though it is probable that normally it remains attached near the middle of the host, sucking its juices through a small puncture made by the constantly moving jaws, seldom or never leaving place, as the skins of the many victims examined show only the one puncture spot. The first larval stage lasts about one day.

The second stage larva, Figure 28, is much larger, especially in head and abdominal region, which have grown faster than

the thorax. The habits are the same, and the duration of this instar is twenty-four hours, on the average.

The third stage larva, Figure 29, is fat and short, measuring as much as 1 by 0.5 mm. when fully fed. The host by this time is very much shrunken but the skin is intact except at the point of puncture. The food of *Lygocerus* is composed of the body liquids of the host, and the internal organs of the latter pulped by the mandibles. The eating process can be easily observed in specimens dissected out of their aphid skins. The mandibles work vigorously all the time and swallowing movements of a paristaltic nature carry the food into the midgut to add to the gluttonous store already there, which because of its yellow color is plainly visible by transmitted light. The third larval stage lasts one and one-half days on the average, but varies a half day each way in some cases.

The fourth stage larva, Figure 30, measures 1.4 by 0.7 mm. The body is tapering, has many short blunt spines or small tubercles on its dorsum and in rows down the sides of each segment, and there is a prominent dorsal projection in the form of a blunt cone on the posterior end of the abdomen. Haviland (1920) has found this in two related European species and suggests a locomotory function. Perhaps this is correct, and yet *Aphidius* larvae living under the same conditions in the same aphid skins are able to move about most actively when spinning cocoons, by the aid of small tubercles such as *Lygocerus* itself possesses, and without the caudal process. Indeed *Lygocerus* need not move about at all, as its cocoon has been spun for it by its victim, and its resting place is all ready for it. During this instar the remains of the host are usually consumed, skin and all; other larvae, seemingly less hungry, leave the host's skin, flattened and sucked dry of contents, and the meconium together as a scale in one end of the skin of the aphid. Just before pupation the *Lygocerus* voids its own meconium, Figures 31 "Mec," which takes the form of a sphere, differing greatly in appearance from the divided meconium of the Primary Parasite. The fourth instar lasts from one and one-half to two days.

There is an interesting kind of peristalsis in the midgut of nearly all larval stages. A broad constriction appears at the anterior end of this part of the digestive system and travels slowly backward to the posterior end, taking about four sec-

onds to reach this point. Then the direction is changed and the constriction moves forward to the anterior end at the same rate, then backward again and so on. The lumen of the constriction is evidently quite small since the particles of food swirl and squirt thru as it moves backward and forward, an unique method of mixing food and digestive juices, possibly resulting in rapid and efficient digestion and absorption.

The pupa, Figure 31, is yellow at first but grows gradually darker until it is black. It lies bent inside the aphid skin, with the ventral surfaces of head and abdomen somewhat approximated. The spherical meconium is found in the space between the abdomen and the head, among the appendages. The abdomen of the pupa, upon stimulation, moves from side to side and back several times much as do certain Lepidopterous pupae, except that the latter twist the abdomen around in a rotary manner. The pupal stage lasts from six to eight days.

A generation of this parasite can be produced in eleven days, and the average time is about twelve days, although a few straggle out to fourteen or sixteen days before emerging.

By dissecting out of the aphid skins just before they were ready to emerge, females were obtained that were unmistakeably virgin. These were placed singly in breeding tubes with numbers of aphids containing *Diaeretus rapae* in the last larval stage. Oviposition was seen to start at once. The progeny, averaging sixteen, were as numerous, as vigorous, and as large as for the mated individuals of the stock series of tubes, but no females were produced, every emerging *Lygocerus* being a male.

Asaphes americana Gir.

In the present work *Asaphes americana* Gir. was found associated with *Diaeretus rapae* Curt. at Raleigh, N. C., in February, 1921. The species also occurred as a hyperparasite on the same host at Columbus, Ohio, and was present until the fall of the year. It reached its greatest abundance in July and August when over fifty-five percent of the parasitized aphids on crucifers bred out this species. After October the numbers dwindled rapidly and during the winter none appeared tho many of the primary parasites were bred from field material. In addition during the past year *Asaphes* has been

bred from aphids parasitized by *Aphidius phorodontis* Ashm., *Aphidius polygonaphis* Fitch, and *Lysiphlebus testaceipes* Cress. Hartley (1922) reports it as a parasite on *Aphelinus semiflavus* How. in the greenhouse during late winter. It is evident that *Asaphes* can reproduce in a number of hosts and one is led to believe, from this list, that aphids parasitized by any of the primary parasites might be subject to attack.

Asaphes emerges from the inflated aphid skin thru an irregular circular hole that differs considerably from the larger, smoothly cut emergence hole of *Aphidius*. Mating occurs as soon as the female is dry, and oviposition as in *Lygocerus* may occur with or without fertilization. For oviposition the female of *Asaphes* hunts an aphid skin containing a late larva or early pupa of *Aphidius* or other primary host. Never is an unparasitized aphid selected or a parasitized one in which the larva has not eaten out the aphid and spun its cocoon. Thus beyond all doubt *Asaphes* is a secondary parasite. Aphid after aphid is investigated by *Asaphes* with her delicate antennae until such a one is found, and then after feeling all around the skin, the little wasp climbs up on the aphid, catches hold with all six legs, squats a little, points her antennae downward and starts drilling with the tip of the ovipositor. It takes all of five minutes constant work for the egg to be placed. The larva develops in an ectoparasitic manner, very much as does *Lygocerus niger*.

To determine the number of eggs laid by one female, and approximately the length of life, a series of experiments were carried on in the laboratory, in all, seventeen different females being studied. The table below gives the results for five females that are representative:

ASAPHES. LENGTH OF LIFE AND NUMBER OF PROGENY.

FEMALE.	DAYS LIVED.	PROGENY.	NOTES.
No. 8231	14	49	A virgin; progeny all males.
No. 9061	11	51	Six females, others of progeny all males.
No. 9062	23	76	A virgin; progeny all males.
No. 9091	14	90	A virgin; progeny all males.
No. 9221	7	5	Two of these females.
Average	14	54	

It will be seen from the above data that each female in her fourteen days of life can parasitize at least fifty-four *Aphidius*, and thus accomplish a great deal of harm by destroying these beneficial insects. The wonder is that any of the *Aphidius* are left with so much in favor of the hyperparasite. However in these experiments indoors a large percentage of males was produced and only a few females, even tho the parent had been fertilized. If this holds for field conditions, the danger from *Asaphes* is lessened. It is interesting to note that no females were among the progeny of virgins. Somehow or other, it seems to be the faculty of nearly every species of aphid parasite and hyperparasite to be able to reproduce parthenogenetically, and in every case but *Aphelinus* the virgin female begets males.

The males can easily be distinguished from females in *Asaphes*, because the former have a much less robust body, and antennae with spatulate tips, whereas the female is larger, possesses an ovipositor under the deeper abdomen, and has the tip of the antenna more conical and tapering. The antenna character is the most dependable, as the size varies with the size of the aphid host in which development takes place.

Pachyneuron apidivorum Ashmead.

Pachyneuron apidivorum Ashm. was bred from *Aphis rumicis* Linn. on September 14, 1921, from nasturtium plants at Columbus, Ohio. The mated females were given young of this aphid and of *Aphis pseudobrassicae* Davis for oviposition but paid no attention to them. However, when placed among aphids that had been previously parasitized by *Lysiphlebus testaceipes* Cress. and *Diaeretus rapae* Curt., whose larvae had just spun their cocoons, no time was lost by the *Pachyneuron* females, which began at once to lay their eggs. Oviposition and development was similar to the processes in *Asaphes*.

The length of time required for the development of egg into adult, from three generations reared during September and October, in *Aphis pseudobrassicae* parasitized by *Diaeretus rapae*, was from twelve to fifteen days, with the majority of the adults appearing on the thirteenth day after oviposition. The fecundity of the females was rather low, the greatest number of eggs laid by one individual being eleven. On ac-

count of this last fact, it is doubtful if *Pachyneuron* will ever play a very great part in the Aphid Complex, unless the habits of the species change greatly under the influence of different conditions that would be more nearly optimum for the parasite. Perhaps it is well that it cannot do more damage than it does.

Xystus brassicæ Ashmead.

In May, 1921, *Xystus brassicae* Ashm. was observed among *Aphis pseudobrassicae* Davis on cabbage plants at Raleigh, N. C., in association with *Asaphes americana* Gir. and *Diaeretus rapae* Curt. Several attempts were made to induce *Xystus* to oviposit directly in *Aphis pseudobrassicae* but without any success, in spite of the fact that many individuals were bred from parasitized aphids of this species from the field, over twenty-five percent of the infested aphids breeding out this species at this time. During the summer at Columbus, Ohio, *Xystus* was also obtained from a number of other aphids, being in each case associated with species of *Aphidiinae*. In late summer it was conceived that *Xystus* might be a hyperparasite on these aphids, developing only upon the developing primary parasites. To test this out two dozen *Xystus brassicae* were introduced into a breeding cage in which *Diaeretus rapae* had been rearing on *Aphis pseudobrassicae* for months. Oviposition in certain of the live aphids was observed at once. It was determined that only those aphids were selected which contained half-grown larvae of *Diaeretus*. Serial sections showed that the larval *Xystus*, (Figures 35, 36, and 37) develops in the body cavity of the larva of *Diaeretus* in a manner entirely similar to the development of the latter in the body of the aphid. Around the larva is a similar serosa, and feeding and respiration occur by osmosis at first, without destruction of the host larva, which is allowed to proceed past the point of cocoon spinning and to pupation before the hyperparasite larva induces destructive changes in the tissues of the primary larva in the same manner as the latter treats the aphid. From this point onward *Xystus* develops at the expense of its host and finally ends by eating the remnants of the latter. It then voids its meconium and pupates, but without spinning a cocoon, since the *Diaeretus* larva has already done this. The adult, Figure 40, eats its way out, leaving an irregular rounded hole, much different from the emergence hole of the *Aphidiinae*.

In the breeding cage mentioned the new generation of *Xystus* appeared in great numbers on November 21, just twenty-six days after the adults were introduced into the cage. The numbers of *Diaeretus* were cut down to about one-tenth, and in another generation disappeared. Then, having been too successful, the *Xystus* parasites perished also.

In the field during the summer of 1921, this parasite was watched with considerable interest and care. It was found most abundant in *Diaeretus rapae*, rather plentiful in *Aphidius polygonaphis* and *Lysiphlebus testaceipes*, and a few times in *Aphidius phorodontis*. A closely related species, *Xystus tritici* Fitch, so much like the species under discussion as to raise doubts about it really being a distinct species, was bred from *Aphidius nigripes* in *Macrosiphum granarium*. The relative abundance of *Xystus brassicae* and its hosts fluctuated considerably during the year of 1921, varying with each variation in number of aphids and parasites, but was always present in numbers great enough to seriously lessen the efficiency of the primary parasites. It was least abundant in the early spring and hot summer months, and most prolific in late spring and late fall thru the winter. The abundance of *Xystus* in pupal condition during the winter was surprising and of importance, as it settled the question of the method of wintering. From one set of twenty-five parasitized *Aphis pseudobrassicae* secured in February at Columbus, there were bred eight *Diaeretus rapae* and sixteen *Xystus brassicae*, a parasitization of two-thirds of the primary parasites. Other lots gave similar results.

INTERRELATIONS.

The relations between the species of hyperparasites, parasites, aphids and plants are complicated in many respects, and difficult to express in a connected manner, but a treatment of the Aphid Parasite Complex would be incomplete without a full discussion of this phase. So a recapitulation of the relations as found is presented in a graphic manner in Text Figure 2.

In this diagram are brought out several significant points. First, the number of important crops in the list of plants infested by aphids of this complex indicates somewhat the economic importance of these forms and their parasites, and justifies

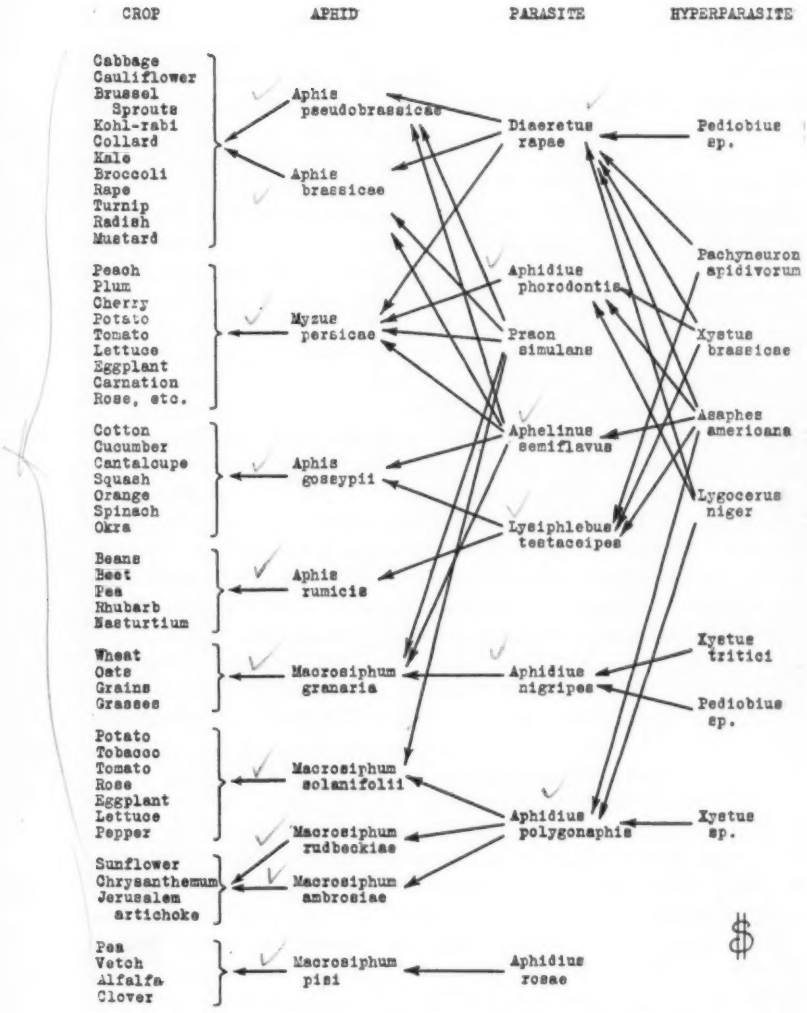


Fig. 2. The Aphid Parasite Complex. Relations between Crops, Aphids, Parasites and Hyperparasites.

a study of the biology of the latter since they are very beneficial. Second, the primary parasites are narrowly restricted to a few aphid hosts in most cases, which is a decided disadvantage from an economic standpoint; but this disadvantage is offset by the abundance of closely related parasite species, so that each one of the pests mentioned has at least one parasite to check its activities. Lastly, the hyperparasites are less restricted as to hosts than the others, perhaps a serious consideration from the economic viewpoint, yet not always so practically, since the life of a hyperparasite is very precarious at best, for if one of the factors upon which it depends drops out or changes very much, it means the end for the hyperparasite.

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EXPLANATION OF PLATES.

PLATE VII.

Development of the *Aphidiinæ*, as illustrated by *Diaeretus rapæ* Curt.

Fig. 1. Ovarian egg. (× 380).
Fig. 2. Egg after oviposition. (× 380).
Fig. 3. Cross-section of ten hour embryo, from serial sections, showing formation of the serosa. (× 380).
Fig. 4. Cross-section of twenty-four hour embryo. (× 380).
Fig. 5. Embryo of forty-eight hours, reconstructed from serial sections. (×150).
Fig. 5a. Cross-section of this embryo. (× 350).
Fig. 6. Reconstruction of embryo of about fifty-five hours. (× 200).
Fig. 7. Embryo of the third day. (× 200).
Fig. 7a. Cross-section through head of this embryo. (× 200).
Fig. 8. First stage larva, reconstructed from sections. (× 50).
Fig. 9. Second stage larva, reconstructed. (× 50).
Fig. 10. Third stage larva, reconstructed. (× 50).
Fig. 11. Fourth stage larva. (× 50).
Fig. 12. The meconium voided by mature larva. (× 50).
Fig. 13. The pupa. (× 50).
Fig. 14. Adult *Diaeretus rapae*. (× 25).

PLATE VIII.

The effects of Parasitism upon various tissues.

Fig. 15A. Surface view of portion of serosa, *Praon simulans*. (× 150).
Fig. 15B. Section through normal serosa, *Praon simulans*. (× 150).
Fig. 16A. Serosal masses after hatching of parasite larva of *Praon simulans* in *M. solaniifolii*. (× 150).
Figs. 16B, 16C, 16D. Masses from later stages, *Praon simulans*. (× 150).
Figs. 17A, 17B, 17C. Similar serosal masses from *Aphis gossypii* parasitized by *Lysiphlebus testaceipes*. (×150).
Figs. 18A, 18B, 18C, 18D. Transformation of similar serosal masses from *Aphis pseudobrassicæ* parasitized by *Diaeretus rapæ*. (×150).
Fig. 19. Normal adipose tissue from *Macrosiphum solanifolii*. (× 800).
Fig. 20. Adipose tissue from an aphid, *Macrosiphum solanifolii* parasitized by *Praon simulans*. (× 800).
Fig. 21. Normal embryo from *Aphis pseudobrassicæ*, sagittal section. (×125).
Fig. 22. Embryo from an *Aphis pseudobrassicæ* parasitized by *Diaeretus rapæ*. (× 125).
Fig. 23. Young larva of *Aphidius polygonaphis* Fitch, from an aphid containing an older larva of the same species. (× 75).
Fig. 24. The normal larva of *Aphidius polygonaphis* Fitch, from same aphid as Fig. 23, and responsible for the degenerate condition of that larva. Reconstructed from sections. (× 75).

PLATE IX.

The development of the Hyperparasite *Lygocerus niger* Howard.

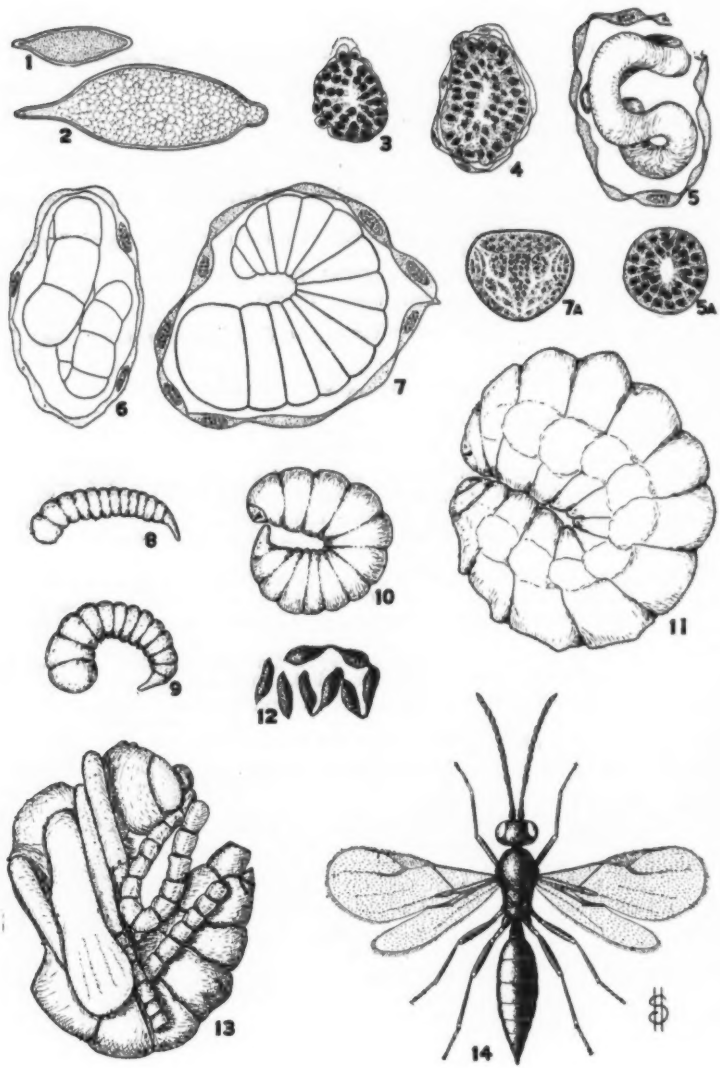
Fig. 25. Egg, dorsal view. (× 75).
Fig. 26. Egg, lateral view. (× 75).
Fig. 27. First stage larva. (× 75).
Fig. 28. Second stage larva (HYP) attacking and eating fourth stage larva of *Diaeretus rapæ* (PAR). (× 37).
Fig. 29. Third stage larva (HYP), attacking and eating pupa of *Diaeretus rapæ* (PAR). (× 37).
Fig. 30. Fourth stage larva (HYP), eating the remains of a pupa of *Diaeretus rapæ* (PAR). (× 37).
Fig. 31. The pupa, with meconium (MEC). (× 37).
Fig. 32. The adult female *Lygocerus niger*. (× 20).

(Figures 25 to 31 are from dissections of parasitized *Aphis pseudobrassicæ*).

PLATE X.

The Parasite *Aphelinus semiflavus* Howard and the Hyperparasite *Xystus brassicæ* Ashmead.

Fig. 33. Cross-section of an *Aphis pseudobrassicæ* containing young larva of *Aphelinus* devouring an aphid embryo. PAR is the larva. (× 85).
Fig. 34. Longitudinal section of a similar aphid containing full-grown larva of *Aphelinus* (PAR), which has devoured nearly all of the aphid's organs. (× 42).
Fig. 35. Egg of *Xystus brassicæ* in unhatched larva of *Diaeretus rapæ*. (× 200).
Fig. 36. Embryonic larva of *Xystus* (HYP) developing in half-grown larva of *Diaeretus*, the latter in body of an *Aphis pseudobrassicæ*. From serial sections. (× 75).
Fig. 37. Half-grown larva of *Xystus* (HYP) in abdomen of full-grown larva of *Diaeretus* (PAR), the latter in aphid. From serial sections. (× 75).
Fig. 38. Mature larva of *Xystus*. Dissected from parasitized *Aphis pseudobrassicæ*. (× 40).
Fig. 39. Pupa of *Xystus*. (× 40).
Fig. 40. Adult female *Xystus brassicæ* Ashm. (× 25).



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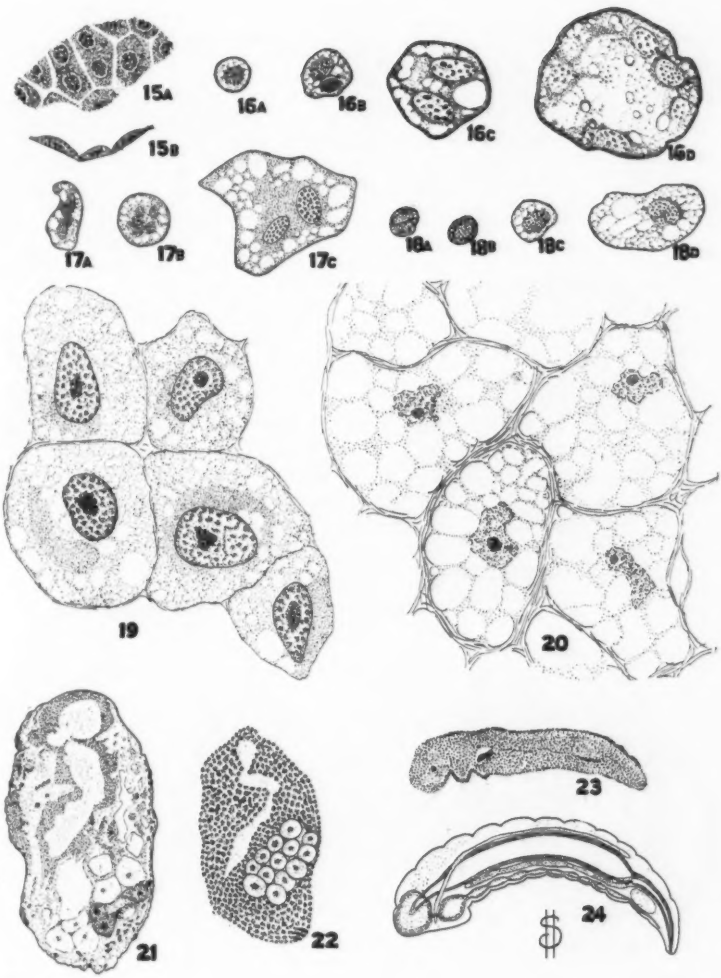

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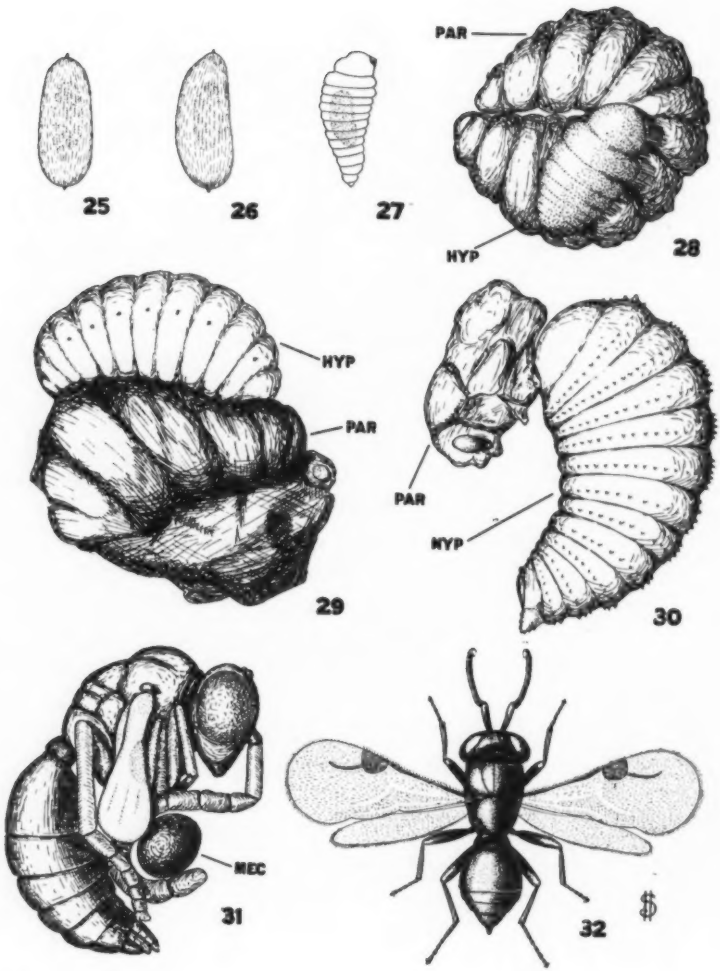

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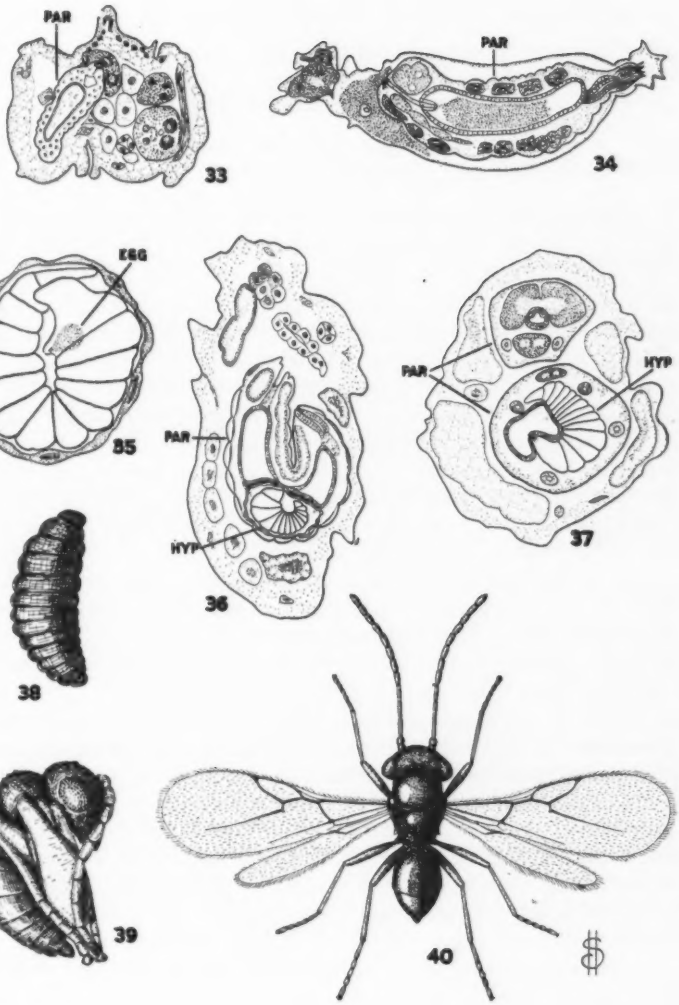

Herbert Spencer

STUDIES ON THE CRANE-FLIES OF MEXICO.

PART II.

(Order Diptera, Superfamily Tipuloidea).

CHARLES P. ALEXANDER,
Amherst, Massachusetts.

The present paper is a continuation of the studies outlined in part I of this series of papers (Annals Ent. Soc. America, 18: 341 – 362; 1925). As before, the species discussed were all included in very large and important series of crane-flies that were collected and sent to me for determination by my friend, Dr. Alfons Dampf, State Entomologist of Mexico. A few brief notes on certain additional *Mexican Biocenosis* (M.B.) and *Mexican Fauna* (M. F.) numbers are given herewith:

Hacienda San Francisco, near *Córdoba:* Swept from the walls of a passage way, cut three to four meters deep across an uncultivated area covered with woods and bushes, the perpendicular walls of the pass overgrown with small ferns and other plants. M.F. 335, 490, etc.

Barranca of the Rio San Antonio, near *Córdoba*, altitude 870 meters, swept from ferns and other plants beneath overhanging trees. The Rio consists of small clear ponds connected by faint water-courses with very stony beds. *Isocoenosis* to M. B. 73 (see Part I, p. 341). M. B. 102.

Cerro de Hongos: Southern prolongation of the Desierto mountain range; swept from grass in open pine forest at an altitude of from 3500 to 3700 meters. M. B. 117.

Ajusco: The highest and most easterly part of the mountain chain south of Mexico City, of which the Desierto forms the western part. Dense *Abies* forest, altitude 3500 meters, swept from weeds along the course of a small brooklet.—*Rhaphidolabis* (*Plectromyia*) *tergata* sp.n. In pine forests between 3500 and 4100 meters.—*Tipula oreomyzoides* sp.n.

I express my deepest thanks to Dr. Dampf for the privilege of studying this splendid series of Mexican Tipulidae, the types of which are retained in my collection through the kindness of the collector.

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*Contribution from the Entomological Laboratory of the Massachusetts Agricultural College.

Limonia Meigen.

Limonia fissilis sp. n.

General coloration brown, the pleura with a conspicuous longitudinal black stripe; rostrum and antennae black throughout; wings with a dusky tinge, the stigma darker; *Sc* and *Rs* long; male hypopygium complicated in structure.

Male. Length about 4.8 mm; wing, 5 mm.

Rostrum and palpi black. Antennae black, the flagellar segments oval. Head dark brown.

Mesonotal praescutum dark brown laterally and more broadly so medially behind, the remainder of the disk more reddish brown; scutal lobes dark brown, the median area and the scutellum more testaceous; postnotum shiny brown. Pleura light brown with a broad black longitudinal stripe extending from the cervical sclerites to the base of the abdomen, passing beneath the root of the halteres; ventral pleurites paler. Halteres dark brown, the base of the stem narrowly paler. Legs with the fore coxae slightly infuscated; remaining coxae and the trochanters yellowish testaceous; femora dark brown, the bases paler; tibiae and tarsi darker brown. Wings (Fig. 1) with a dusky tinge, the stigma oval, darker brown; slightly paler brown clouds at origin of *Rs*, tip of *Sc* and as vague seams along the cord and outer end of cell *1st* M_2; veins dark brown. Venation: *Sc* long, Sc_1 extending to opposite two-thirds the length of *Rs*, Sc_2 at its tip; *Rs* long, gently arcuated; cell *1st* M_2 large, widened distally; *m-cu* close to the fork of *M*.

Abdominal tergites dark brown, the sternites vaguely bicolorous, the bases being paler than the tips. Male hypopygium (Fig. 12) with the ninth tergite (t) nearly straight across the caudal margin, with a large patch of setae on either side. Basistyle (b) relatively large, the ventro-mesal lobe very large and complicated in structure; on its outer side near base bearing a small, densely hairy, finger-like lobule; lobe dilated outwardly, the inner margin before apex dilated into a ledge that is microscopically setulose; apex of lobe terminating in a small conical papilla. Dorsal dististyle (dd) boomerang-shaped. Ventral dististyle (vd) very complex, the rostral prolongation large. Phallosome (p) forming a large central mass.

Holotype, ♂, Tres Valles, Vera Cruz, September 26, 1924; in hotel (A. M. Dampf).

Rhipidia Meigen

Rhipidia (Rhipidia) **multifida** sp. n.

Male with ten branched flagellar segments; flagellum bicolorous; thorax anteriorly obscure yellow, the posterior sclerites of the notum and pleura darkened; all tarsi infuscated; wings tinged with dusky, with three additional smaller dark spots and inconspicuous seams along the cord; *m-cu* before the fork of *M;* male hypopygium with the dorsal dististyle long and slender, strongly curved near midlength.

Male. Length about 5 mm.; wing, 5.8 mm.

Rostrum and palpi black. Antennae with the scapal segments black; basal enlargements of the flagellar segments and the branches black, the elongate pedicel of the individual segments whitish; ten branched flagellar segments, the longest branch about three times the length of the segment; first flagellar segment with a conically produced apical projection. Head dark brownish gray. Eyes (♂) almost touching, the anterior vertex being reduced to a narrow strip.

Pronotum dark brown. Mesonotal praescutum obscure yellow, with a brown median stripe that is slightly interrupted near midlength; lateral stripes very short and paler in color; scutum entirely dark brown; scutellum and postnotum slightly paler brown. Pleura obscure yellow, the posterior pleurites (pteropleurite, meron and all sclerites caudad of them) infuscated. Halteres obscure yellow, the knobs infuscated. Legs with the fore coxae obscure yellow; middle and hind coxae dark brown; trochanters brownish testaceous; femora brown, the bases somewhat paler; tibiae and tarsi brown, the terminal tarsal segments brownish black. Wings (Fig. 2) with a strong dusky tinge, the stigma oval, slightly darker brown, the outer end darker than the remainder and appearing as a seam at *r;* small brown clouds at origin of *Rs* and tip of *Sc;* very narrow and vague dusky seams along cord and outer end of cell *1st* M_2; prearcular cells pale; veins dark brown. Venation: *Sc* relatively short, Sc_1 ending just before midlength of *Rs*, Sc_2 at its tip; *Rs* long, weakly angulated at origin; *m-cu* shortly before the fork of *M; h* apparently lacking.

Abdominal tergites dark brown; sternites obscure yellow, the caudal margins of the segments narrowly ringed with brownish black. Male hypopygium (Fig. 13) having the ninth tergite with a deep U-shaped notch caudally, the lateral lobes conspicuous, fringed with powerful spinous bristles. Basistyle (b) relatively small, the ventro-mesal lobe narrowed outwardly, provided with numerous tubercles that bear conspicuous setae, some of which are exceedingly elongate. Dorsal dististyle (dd) long and slender, at midlength bent at about a right angle, near the apex suddenly narrowed into a long, acute, blackened spine. Ventral dististyle (vd) fleshy, larger than the basistyle, the rostrum elongate; rostral spines two, the outer one much stouter (its tip broken and total length not certain), the inner spine more slender, setiform. Gonapophyses (g) conspicuously blackened. Aedeagus (a) at apex split into two lobes.

Holotype, ♂, Córdoba, September 30, 1924 (A. M. Dampf); M. F. No. 385.

Rhipidia (Arhipidia) **agglomerata** sp. n.

Male. Length 4.5—4.7 mm; wing, 6.2—6.3 mm.

Closely allied to *R.* (*A.*) *multiguttata* Alexander, of Guatemala, to which species it runs by means of the author's key to the New World species of *Rhipidia* (Bull. Brooklyn Ent. Soc., 8: 7-8; 1912). The present species differs as follows:

Head brownish gray. Mesonotal praescutum rich reddish brown in front, the three brownish black stripes indicated behind, crossing the suture and suffusing the scutal lobes; scutellum pruinose; postnotal mediotergite baso-medially brownish black. Ventral brown pleural stripe broader and darker, suffusing the basal halves of the middle and posterior coxae. Legs with the femora obscure yellow, and tips a little clearer yellow but the segments without distinct darker subterminal rings. Wings with the brown dots in the cells of the wings relatively few in number, tending to mass into irregular blotches. Venation: Sc_1 ending opposite or beyond midlength of *Rs*.

Holotype, ♂, Córdoba, Rio San Antonio, after sunset, September 30, 1924 (A. M. Dampf); M. F. 385.

Paratopotypes, 2 ♂ ♂.

The hypopygial details in these two species are very similar to one another. The same general type is found not only in other species of *Arhipidia*, but in the typical subgenus (*R. longispina* Alexander.) One of the paratypes shows a remarkable deformity of the male hypopygium, the dististyles of the right side being duplicated; the basistyle of this side is larger than usual. The gonapophyses and aedeagus likewise appear to be asymmetrically repeated.

Helius St. Fargeau.

Helius quadrifidus sp. n.

Allied to *H. albitarsis* (O. S.); size small (wing, ♂, 4 mm.); wings with a dusky tinge, veins R_{4+5} and M_{1+2} very extensively fused, obliterating *r-m* and giving the appearance of four veins issuing from cell *1st* M_2.

Male. Length, 4 mm; wing, 4 mm.

Rostrum and palpi black. Antennae black. Head black.

Thorax dark brown, the mesopleura paler brown. Halteres dark brown. Legs with the coxae and trochanters dark brown; femora and tibiae black, the extreme bases of the former somewhat paler; tarsi black, the end of the basitarsus and all the remaining tarsal segments except the last pure white, clothed with snowy white setae. Wings (Fig. 3) with a dusky tinge, the oval stigma somewhat darker; veins dark brown. Venation: Sc_1 ending opposite three-fourths the length of *Rs*, Sc_2 at its tip; *Rs* nearly straight; R_{2+3} sinuous; R_{4+5} very extensively fused with M_{1+2} to form the cephalic margin of cell *1st* M_2, giving the appearance of four veins issuing from the cell; basal deflection of R_{4+5} short; cell *1st* M_2 very large; *m-cu* shortly beyond midlength of the cell.

Abdomen black, the basal sternites paler; hypopygium black.

Holotype, ♂, At light on train between La Favorita and Tapachula, State of Chiapas, June 14, 1925 (A. M. Dampf); M. F. No. 558.

Orimarga Osten Sacken.

Orimarga dampfi sp. n.

General coloration gray, the mesonotal praescutum with a median brown stripe; thoracic pleura with a conspicuous silvery white stripe; legs pale brown; wings with a pale grayish tinge, the base whitish; no macrorichiae on distal section of vein M_4.

Male. Length, 7.5–8 mm.; wing, 5.5 mm.

Female. Length about 8.5 mm.; wing, 6–6.2 mm.

Rostrum and palpi brownish black. Antennae dark brown, pruinose; flagellar segments globular to short-oval. Head light gray.

Mesonotum dark gray, the praescutum with a median brown stripe, the extreme lateral margins of the sclerite pale. Pleura brownish gray, with a conspicuous silvery white stripe extending from behind the fore coxa, across the dorsal edge of the sternopleurite, ending above the posterior coxa. Halteres short, pale yellow. Legs with the coxae brownish testaceous, the trochanters obscure yellowish brown; femora pale brown, the remainder of the legs somewhat darker brown, the tips of the individual segments narrowly darker, the terminal tarsal segments more uniformly darkened. Wings (Fig. 4) with a very pale gray tinge, the prearcular region whitish; veins pale brown. Macrotrichiae on veins rather numerous, there being about 20 on the distal section of vein M_3, about 24 on the distal section of R_{2+3}; no macrotrichiae on M_4. Venation: *Sc* ending opposite one-third the length of *Rs*, Sc_2 at its extreme tip; *Rs* gently angulated at origin; *r* about one and one-half times its length from the tip of R_1 and about its length beyond the fork of *Rs*; *r* near midlength of the basal section of R_{4+5}; M_4 a little more than one-half the distal section of M_3; *m-cu* oblique, near midlength of *Rs*.

Abdomen elongate, dark grayish brown.

Holotype, ♂, Vera Cruz, November 10, 1923 (A. M. Dampf).
Allotopotype, ♀.
Paratopotypes, 30 ♂ ♀.

This interesting species is named in honor of the collector, Dr. Alfons Dampf, to whom we are indebted for much of our knowledge of the crane-flies of Mexico.

Erioptera Meigen.

Erioptera (Mesocyphona) **venustipes** sp. n.

Belongs to the *dulcis* group; head variegated; fore and middle femora each with three broad black rings; posterior femora black with a narrow yellow subterminal ring; tips of the tibiae narrowly brownish black.

Female. Length about 3.5 mm.; wing, 4.8 mm.

Rostrum and palpi black. Antennae with the scapal segments brownish black, the basal flagellar segments pale yellow, the terminal

segments passing into pale brown. Head dark brown, the orbits narrowly but conspicuously yellowish.

Lateral pretergites narrowly pale sulphur-yellow. Mesonotal praescutum brownish gray with two conspicuous dark brown stripes that are subequal in width to the space separating them, these dark stripes continued caudad onto the scutal lobes; lateral margins of the praescutum passing into light brown, the extreme lateral edge dark brown; pseudosutural foveae pale; scutellum with the caudal margin broadly yellow. Pleura dark brown and silvery, alternated, the dark brown including the dorsal half of the dorso-pleural region; a conspicuous stripe extending from and including the fore coxa, passing above the halteres to the postnotum; a narrow ventral stripe extending from the sternopleurite passing above the middle coxa and below the halteres to the abdomen; a conspicuous sulphur-yellow stripe occupies the ventral portion of the dorso-pleural membrane, extending from above the fore coxa to the wing-root; the silvery stripes remaining are two in number, the dorsal one narrowed in front, widening behind and including the base of the halteres; the ventral silvery stripe includes the broad meron and the katepisternum. Halteres white, the knobs infuscated. Legs with the fore coxae dark brown; middle and hind coxae pale, the apices of each with a dark spot; trochanters pale; fore and middle femora yellow, each with three conspicuous brownish black annuli that are much more extensive than the ground-color, the first pre-medial, the second, a little wider, post-medial, the third broadest, subapical; posterior femora entirely brownish black except for a conspicuous yellowish white ring far before the tip and the extreme apex of the segment which is pale; tibiae yellowish white, the tips narrowly brownish black; two basal segments of tarsi yellowish white, the tips and the remainder of the tarsal segments dark brown. Wings with a brown suffusion, more intense apically and in the radial field; basal half of cells *C* and *Sc* more yellowish; prearcular cells dark; disk with twenty large white spots arranged about as in the other members of the *dulcis* group, including spots at *h*, Sc_2, Sc_1, along the cord and fork of M_{1+2}; a series of nine marginal spots between veins R_1 and *1st A;* the remaining spots are located at arculus, origin of *Rs*, on *Rs* just before the fork; on M_{1+2} shortly beyond *m;* and two areas along vein *1st A*, one near the base, the second near midlength of the vein; anal angle restrictedly pale; veins brown, in the pale areas whitish; vein *R* largely black, R_1 and Cu_1 largely yellow. Venation: Cell *1st* M_2 open by the atrophy of the outer deflection of M_3.

Abdominal tergites brown, the lateral margins of the segments narrowly blackened; sternites more yellowish brown, with numerous brown setigerous punctures.

Holotype, ♀, Coscomatepec, altitude 1580 meters, flying over the surface of the Rio Tlacuapan, a swift-flowing stream with sandy bottom and very clear water, November 29, 1924 (A. M. Dampf); M. F. No. 463.

Paratopotype, ♀ ; M. F. No. 461.

E. venustipes belongs to the *dulcis* group of the subgenus, which now includes *dulcis* Osten Sacken, *eiseni* Alexander, *needhami* Alexander, and the present species.

Gonomyia Meigen.

Gonomyia (Gonomyia) **remota** sp. n.

Belongs to the *subcinerea* group; antennal scape largely yellow; rostrum black; head yellow with a dark spot on vertex; pleura yellow with two dark brown stripes; wings with *Sc* short, Sc_1 ending far before the origin of *Rs*, Sc_2 lying detached, some distance distad of Sc_1; abdominal tergites dark brown, with conspicuous caudo-lateral yellow triangles.

Sex? Length about 3.8 mm.; wing, 4.6 mm.

Rostrum and palpi black. Antennal scape conspicuously yellow with vague darker markings; flagellum dark brown; flagellar segments oval, passing into elongate-oval. Head light yellow, the center of the vertex with a dark brown spot.

Pronotum light sulphur yellow. Lateral pretergites similar. Mesonotal praescutum brown with three nearly confluent darker brown stripes, the surface weakly pruinose; lateral margins of sclerite and the humeral triangle obscure brownish yellow; pseudosutural foveae elongate, reddish brown; scutum obscure yellow, the lobes extensively dark brown, confluent across the suture with the lateral praescutal stripes; scutellum testaceous yellow; postnotal mediotergite yellow, darkened posteriorly. Pleura yellow, with two conspicuous dark brown stripes, the dorsal one extending from the cervical sclerites, passing above the root of the halteres and including the posterior margin of the postnotum; ventral stripe including the stenopleurite and meron. Halteres slender, dark-colored. Legs with the coxae and trochanters light yellow; remainder of legs dark brown. Wings (Fig. 5) with a faint brown tinge, the stigma slightly darker; veins darker brown. Venation: *Sc* very short for a member of the *subcinerea* group, Sc_1 ending a distance before the origin of *Rs* that is nearly as long as the latter; the detached Sc_2 lies considerably beyond the end of Sc_1 at near two-fifths the distance between Sc_1 and the origin of *Rs; Rs* strongly arcuated, about equal to the slightly less arcuated R_{2+3}; basal deflection of R_{4+5} short; *m-cu* immediately beyond the fork of *M*.

Abdominal tergites dark brown; tergites two to four with conspicuous light yellow caudo-lateral triangles; abdomen broken beyond the fourth segment; sternites light yellow.

Holotype, Sex? , Barranca of the Rio San Antonio, near Córdoba, altitude 870 meters, November 20, 1924 (A. M. Dampf); M. B. No. 102.

Gonomyia remota departs from the normal characters of the *subcinerea* group in the yellow antennal scape and short subcosta. The detached Sc_2 is unique in the genus, as far as known.

Gonomyia (Lipophleps) **strigilis** sp. n.

Male. Length about 3.2 mm.; wing, 3.8 mm.

Belongs to the *cinerea* group; most closely allied to *G.* (*L.*) *cinerea* (Doane), differing chiefly in the structure of the male hypopygium.

Rostrum and palpi black. Scapal segments of antennae brownish yellow, blackened beneath. Head yellow, the center of the vertex infuscated. Mesonotal praescutum brown, the lateral margins paler; pseudosutural foveae conspicuous, reddish brown; posterior sclerites of mesonotum weakly pruinose; a capillary median vitta extending from the suture to the abdomen. Pleura with the whitish longitudinal stripe fully twice as wide as the brown stripes that enclose it. Wings with the costal margin china-white as in the group. Male hypopygium (Fig. 14) with the ventral or outer dististyle (d) a long blade, the apical half infuscated, on the mesal face at near the basal third with a small, subappressed black spine, the margins of the style not otherwise armed; intermediate dististyle a short blackened spine, narrowed to the acute apex; dorsal or inner style a pale fleshy lobe, the caudal margin produced caudad into a blackened spine.

Holotype, ♂, Between Coscomatepec and Tomatlan, altitude nearly 1500 meters, swept from the train, from grass and bushes, November 30, 1924 (A. M. Dampf); M. F. No. 466.

The chief differences between the present species and *G. cinerea* are the small spine on the ventral dististyle and the short, heavily blackened intermediate dististyle.

Gonomyia (Lipophleps) **latistyla** sp. n.

Belongs to the *pleuralis* group; size small (wing, ♂, less than 3.5 mm.); male hypopygium with the dististyle appearing as a flattened blade, the lateral apical angle produced into a small spinous lobe, the apex of the style with numerous setae and a conical spine.

Male. Length about 3.5 mm.; wing, 3.2–3.3 mm.

Rostrum and palpi black. Antennal scape obscure orange; flagellum black. Head pale orange-yellow.

Praescutum pale brown, the lateral margins broadly light yellow, margined internally by a narrow dark brown line; scutal lobes pale brown; scutellum darker brown; postnotal mediotergite brownish yellow, indistinctly variegated medially with darker. Pleura brown, with a conspicuous white longitudinal stripe extending from the bases of the fore coxae, across the anepisternum and ventral margin of the pteropleurite to the base of the abdomen; this stripe is margined both dorsally and ventrally by a slightly narrower dark brown line; sterno-

pleurite and meron pale. Halteres pale brown, the knobs a little infuscated. Legs with the coxae pale, the apical half of the fore coxa dark brown; femora brown, the tips a little darker; remainder of legs dark brown. Wings tinged with dusky, the oval stigma dark brown; the region before and beyond the stigma paler; base of wing and costal region slightly more yellowish; veins pale, the cord darker. Venation: *Sc* short, Sc_1 ending a distance before the origin of *Rs* that is about two-thirds the length of the latter vein; R_{2+3} short and arcuated, bending around the stigma; cell *1st* M_2 strongly narrowed at proximal end; *m-cu* shortly before fork of *M*.

Abdomen dark brown. Male hypopgium (Fig. 15) of the general type of *G.* (*L.*) *pleuralis* (Will.); chitinized spines at apex of basistyle (b) shorter, the longer slender, curved, the tip acute, the base not heavily chitinized; the shorter spine is very small and pale throughout. Dististyle (d) a broad flattened blade, the outer apical angle produced into a small spinous lobe, the terminal surface of the style with numerous setae and a conspicuous conical spine at the inner apical angle; outer margin of style with a pale flange.

Holotype, ♂, Vera Cruz, Mexico, November 10, 1923 (A. M. Dampf).

Paratopotype, ♂; *paratype*, ♂, Tempisque, Costa Rica, January 18, 1921 (A. Alfaro).

Gonomyia (Lipophleps) **spinistyla** sp. n.

Male. Length about 3 mm.; wing, 3 mm.

Belongs to the *pleuralis* group; most closely allied to *G.* (*L.*) *latistyla* sp.n., differing especially in the structure of the male hypopygium.

Mesonotum rich reddish brown, the scutal lobes darker; median region of scutum yellow with a narrow dusky vitta that extends caudad onto the median area of the scutellum, the latter broadly light yellow behind; postnotum dark at base, the remainder more yellowish. Pleural stripe creamy-white. Abdomen with the disk of the segments pale, the lateral margins broadly darker. Male hypopygium (Fig. 16) with the apical spine of basistyle (b) relatively short and stout, cylindrical, the tip suddenly narrowed into an acute point; ventral fleshy lobe of basistyle moderately stout; the second chitinized spine of the basistyle is apparently lacking in the species. Dististyle (d) pale, the outer margin armed with an acute black spine, the apex of the style beyond this spine narrowed and provided with erect setae, the two apical ones much larger. Phallosome asymmetrical, as figured.

Holotype, ♂, At light on train, between La Favorita and and Tapachula, State of Chiapas, June 14, 1925 (A. M. Dampf); M. F. No. 558.

Gonomyia (Lipophleps) **bifida** sp. n.

Male. Length about 4 mm.; wing, 3.6 mm.

Female. Length about 4.5 mm.; wing, 4 mm.

Belongs to the *pleuralis* group; most closely allied to *G.* (*L.*) *recurvata* Alex.

Rostrum and palpi black. Basal antennal segments orange, this coloration including the first segment of flagellum; remainder of organ dark brown. Head light yellow, the center of the vertex broadly infuscated.

Pronotum sulphur-yellow. Mesonotal praescutum rich reddish brown, the lateral margin narrowly light yellow, the color margined internally by a narrow brownish black line that ends before the cephalic end of the sclerite which remains of a rich reddish brown; a median dark stripe that is better indicated posteriorly; scutal lobes reddish brown, the median area broadly pale with a capillary brown median vitta that passes caudad onto the testaceous yellow scutellum; postnotum pale, dark at the cephalic portion, the caudal end slightly darkened. Pleura light yellow with two narrow brownish black stripes, the space between light yellow and a little wider; dorso-pleural area more chestnut brown.

Abdominal tergites obscure yellow, the caudal and lateral margins of the segments darker, each segment with a narrow, dark, longitudinal median stripe that is more expanded anteriorly. Male hypopygium (Fig. 17) with the outer angle of the basistyle (b) prolonged into a gently curved acicular spine; outer fleshy lobe moderately stout. Dististyle (d) profoundly bifid, the lateral arm a slender, gently curved spine, the inner arm paler, the apex terminating in two powerful bristles, the surface of this arm with numerous smaller setae; at extreme base of dististyle a tiny chitinized peg or spine. Gonapophyses (g) with the blades elongate, acicular, divergent; ventral gonapophyses (g) very short, about as broad as long, the apex terminating in two small, divergent spines.

Holotype, ♂, Tospan, near Córdoba, swept from walls of a way, December 15, 1925 (A. M. Dampf); M. F. No. 490.

Allotopotype, ♂, with the type and pinned on same pin.

Paratype, ♀, San Francisco, near Córdoba, December 10, 1924 (A. M. Dampf); M. F. 474.

Gonomyia (Lipophleps) **haploa** sp. n.

Male. Length about 3.3. mm.; wing, 3.7 mm.

Belongs to the *manca* group; resembling *G.* (*L.*) *inermis* Alex., differing especially in the structure of the male hypopygium.

Rostrum obscure orange; palpi black. Antennae black throughout, the verticils very elongate. Head black, the center of the vertex with an orange-yellow spot.

Pronotum dark. Mesonotal praescutum shiny, rather dark brown, the pretergites conspicuously pale whitish yellow, this coloration ex-

tending to the wing-root; pseudosutural foveae shiny brown; scutellum broadly yellow, with a dark brown median basal spot. Pleura dusky, sparsely pruinose, with a broad white longitudinal stripe extending from the fore coxa, beneath the root of the halteres, to the abdomen. Halteres with the knobs dark. Legs with the coxae and trochanters brownish testaceous; remainder of legs darker brown. Wings with a grayish tinge, the stigma lacking; veins dark brown.

Abdominal tergites dark brown. Male hypopygium (Fig. 18) with the basistyle (b) long and slender, narrowed distally, the surface with setae, including a longitudinal series of stouter setae along the mesal face. Dististyle (d) single, simple, entirely pale, appearing as a direct apical prolongation of the basistyle, the apex with two powerful setae, the surface with about six smaller setae, the outer surface with abundant erect setulae. Phallosome (p) pale, asymmetrical, the longest element (aedeagus, a) long and slender, projecting far beyond the other elements of the hypopygium, the gonapophyses small and only weakly chitinized.

Holotype, ♂, Barranca of the Rio San Antonio, near Córdoba, altitude 870 meters, November 20, 1924 (A. M. Dampf); M. B. No. 102.

The members of the *manca* group in which the male hypopygium has but a single, entirely fleshy dististyle are: *manca* O.S., *inermis* Alex., *phoroctenia* Alex. and the present form. The species are readily separated from one another by the details of structure of the phallosome.

Teucholabis Osten Sacken.

Teucholabis minuta sp. n.

General coloration yellow, the praescutum with three reddish brown stripes, the scutal lobes darker; pleura yellow with a darker dorsal longitudinal stripe; knobs of halteres dark brown; wings hyaline, the stigma lacking; *Sc* short; cell *2nd* R_1 at wing-margin much narrower than cell R_3.

Male. Length about 3 mm.; wing, 3.1 mm.

Rostrum and palpi dark, the latter slender. Antennae dark brown throughout, the flagellar segments with relatively long verticils. Head light gray.

Pronotum yellow. Mesonotal praescutum sulphur-yellow, with three reddish brown stripes, the median stripe darker anteriorly, the lateral stripes crossing the suture and suffusing the scutal lobes; scutellum yellow; postnotum with the mediotergite infuscate, the posterior third brighter. Pleura whitish yellow, including the pleurotergite, the dorsal sclerites suffused with brown to produce a longitudinal stripe. Halteres testaceous, the knobs dark brown. Legs with the coxae and trochanters yellow; femora obscure yellow, the tips conspicuously though narrowly blackened, the amount subequal on all the

legs; tibiae brownish yellow, the tips passing into black; basal half of basitarsi pale, the remainder of the tarsi passing into black. Wings (Fig. 6) hyaline, the stigma lacking; veins dark brown. Macrotrichiae of veins relatively sparse, there being only three on the distal section of R_{2+3}, widely scattered along the vein. Venation: Sc_1 ending shortly beyond the origin of Rs; r almost at the tip of R_1, the latter being about one-third the vein; cell R_3 at wing-margin nearly twice as wide as cell *2nd* R_1; m a trifle shorter than the outer deflection of M_3.

Abdomen brown, the sternites paler; hypopygium dark brown. Male hypopygium of the general structure of *T. pabulatoria* Alexander, but the short dististyles of different shape, the outer arm not extended into a long point.

Holotype, ♂, Vera Cruz, November 10, 1923 (A. M. Dampf); along margins of a ditch.

T. minuta is the smallest species of the genus yet described. Its nearest ally is *T. pabulatoria* Alexander, of Guatemala.

Pilaria Sintenis.

Pilaria rubella sp. n.

Antennae of male elongate; general coloration of thorax shiny reddish brown; wings with a strong brownish tinge, the oval stigma slightly darker brown.

Male. Length, 7–7.5 mm.; wing, 8–8.8 mm.

Rostrum ferruginous, the terminal segments of the palpi dark brown. Antennae with the scapal segments orange-brown; flagellum brownish black, the segments with a dense erect pubescence, in addition to the elongate verticils. Head with the center of the vertex dark brown, the color passing into reddish brown on the genae.

Thorax shiny reddish brown to ferruginous, without evident darker markings, only the scutal lobes somewhat darker, the pale median area of the scutum continued cephalad across the suture onto the praescutum. Halteres brown, the knobs darker. Legs with the coxae and trochanters reddish brown; femora yellowish brown with darker brown setae; tibiae brown, the tips and the tarsi passing into brownish black. Wings (Fig. 7) with a strong brown tinge, the oval stigma setiferous and slightly darker brown; veins dark brown. Venation: Sc_1 ending about opposite two-thirds the length of Rs, Sc_2 at its tip; r immediately beyond the fork of R_{2+3} and a little more than twice its length from the tip of R_1; R_2 relatively short, arched near midlength; inner ends of cells R_3, R_5 and *1st* M_2 in oblique alignment, cell *1st* M_2 lying more basad than the other elements; *m-cu* just before to just beyond midlength of cell *1st* M_2; cell M_1 longer than its petiole.

Abdominal tergites dark brown, indistinctly variegated with paler; sternites obscure brownish yellow; hypopygium reddish yellow.

Holotype, ♂, Córdoba, Hacienda San Francisco, altitude 900 meters; swept from perpendicular walls of a passage way

that was covered with small ferns and other vegetation; September 13, 1924 (A. M. Dampf); M. F. No. 335.

Paratopotype, ♂.

P. rubella differs from *P. tenuipes* (Say) in the coloration of the thorax and the strongly infumed wings.

Atarba Osten Sacken.

Atarba mexicana sp. n.

General coloration shiny brownish yellow; antennae bicolorous, the flagellar segments black, the apices of the segments narrowly pale yellow; wings with veins *Sc* and *Rs* elongate; cell *1st* M_2 closed; abdomen bicolorous, the basal portion of the segments black, the apical portion broadly brownish yellow.

Male. Length about 4.5 mm.; wing, 6 mm.

Rostrum obscure brownish yellow; palpi black. Antennae elongate, if bent backward extending about to the third abdominal segment; scapal segments dark brown; flagellar segments black, the apex of the segments light yellow, that of the first segment including about one-sixth the total length, of the second and third segments including approximately one-third the length; on the succeeding flagellar segments the amount of yellow becomes gradually less, the terminal four or five segments uniformly dark. Head dark brown.

Mesonotum shiny brownish yellow, without distinct markings. Pleura somewhat more yellowish, the dorso-pleural region darker brown. Halteres obscure yellow, the knobs dark brown. Legs with the coxae and trochanters yellowish testaceous; femora yellow, the tips very narrowly and vaguely darkened; tibiae obscure yellow; tarsi passing into dark brown. Wings (Fig. 8) with a strong brownish yellow tinge, the costal region somewhat brighter yellow, especially in the stigmal region; veins brown, those in the costal region paler, especially *Sc*. Venation: *Sc* long, Sc_1 ending beyond four-fifths the length of *Rs*, Sc_2 some distance from its tip; *Rs* strongly angulated and weakly spurred at origin; cell R_1 at wing-margin nearly three times as wide as cell R_3; cell *1st* M_2 closed, rectangular, *m-cu* beyond mid-length.

Abdomen bicolorous, the basal half or less of the segments blackened, the apical portion broadly brownish yellow; hypopygium obscure yellow; sternites somewhat less distinctly bicolorous. Male hypopygium with the aedeagus very elongate, decurved.

Holotype, ♂. Córdoba, near the Rio San Antonio, October 8, 1924 (A. M. Dampf); M.F. No. 401.

Atarba aperta sp. n.

Female. Length about 5 mm.; wing, 5.6 mm.

Generally similar to *A. mexicana* sp.n., differing especially in the venation, trichiation and the coloration of the antennae and abdomen.

Antennae broken near mid-length, if complete extending approximately to the base of the abdomen; flagellar segments uniformly pale brown. Head brown.

Pleura slightly pruinose. Wings (Fig. 9) with cell *1st* M_2 open by the atrophy of the outer deflection of M_3; Sc_1 shorter than in *mexicana*. Macrotrichiae of wing-veins less abundant than in *mexicana*, the bases of the longitudinal veins largely without trichiae, this glabrous area including the basal fourth of *Sc*, the basal three-fourths of *M* and the basal two-thirds of Cu_1. In *mexicana*, the trichiae on *Sc* extend basad almost to arculus; occupy the distal four-fifths of *M;* are much more numerous on the Anal veins, especially *1st A*.

Abdominal tergites dark brown, at most vaguely bicolorous; basal sternites obscure yellow. Genital segment obscure yellow, the valves of the ovipositor horn-yellow.

Holotype, ♀, Heights of Penna Blanca, a mountain chain north of Córdoba, white limestone cliffs in dense forest, swept from sunny places, altitude 1200 meters, December 15, 1924 (A. M. Dampf); M.B. 110.

A Key to the American Species of Atarba.

1. *Sc* long, Sc_1 ending beyond midlength of the long *Rs* 2
   Sc short, Sc_1 ending approximately opposite the origin of the short *Rs* 3
2. Cell *1st* M_2 open; vein *M* with macrotrichiæ confined to distal fourth. (Mexico) ..... *aperta* sp. n.
   Cell *1st* M_2 closed; vein *M* with macrotrichiæ almost to base. (Mexico), *mexicana* sp. n.
3. Femora with a brown subapical ring; wings with a vague darker marking along the cord 4
   Femora without a subapical ring; wings unmarked, except for the stigma ... 5
4. Mesonotum shiny yellow, the praescutum with a brown median stripe, each scutal lobe with two dark brown spots; wings with a narrow brown seam along cord and outer end of cell *1st* M_2. (Paraguay), *punctiscuta* Alex.
   Mesonotum yellowish, unmarked; wings with a dusky spot on anterior cord. (Colombia) ..... *columbiana* Alex.
5. Antennal flagellum uniformly brownish black. (Colombia), *brunneicornis* Alex.
   Antennal flagellum bicolorous; (in *megaphallus* very indistinctly so, only the incisures between the segments being paler) 6
6. Flagellar segments dark brown, with both ends very narrowly and indistinctly pale; male hypopygium with the aedaegus greatly dilated. (Brazil) ..... *megaphallus* Alex.
   Flagellar segments with one end broadly pale; male hypopygium with the aedeagus of moderate size only 7
7. Flagellar segments dark brown or black with the apical portion conspicuously yellow. (British Guiana) ..... *cincticornis* Alex.
   Flagellar segments dark brown or black with the basal portion conspicuously yellow 8
8. Femora with the tips narrowly blackened. (Paraguay) ..... *fiebrigi* Alex.
   Femora uniformly yellowish 9
9. Abdominal tergites dull yellow; size larger (wing, ♀, over 5.5 mm.) (Eastern North America) ..... *picticornis* O. S.
   Abdominal tergites dark brown; size small (wing, ♀, under 5 mm.) (Peru), *varicornis* Alex.

Eriocera Macquart.

Eriocera pallidipes sp. n.

General coloration brown, the pleura largely black; tips of the tibiae and all but the apices of the tarsi obscure brownish white; wings with a pale brown tinge, the costal region and most of the longitudinal veins clouded with darker brown; *Rs* long, cell M_1 lacking.

Male. Length, 12 mm.; wing, 10–10.4 mm.

Female. Length, 14 mm.; wing, 10.2–10.5 mm.

Rostrum and palpi black. Antennae short, the scapal segments obscure brownish yellow; flagellum black, the first segment paler at base; flagellar segments elongate, gradually shortening outwardly. Head dark plumbeous; vertical tubercle small.

Mesonotum brown, the praescutum behind, the scutal lobes and the scutellum more plumbeous. Pleura largely black, the coloration including the postnotal pleurotergite; sternopleurite and meron paling into shiny brownish yellow. Halteres relatively short, the knobs dark brown. Legs with the coxae shiny obscure yellow, the extreme bases of the fore and middle coxae blackened; trochanters yellow; femora obscure yellow, the tips narrowly blackened; tibiae brown, the tips narrowly whitened; tarsi obscure brownish white; the terminal segments passing into brown. Wings (Fig. 10) with a pale brownish tinge, the stigma scarcely indicated, the base, costal region and seams along most of the veins darker brown; veins dark brown. Venation: Sc_1 ending about opposite two-thirds R_{2+3}, Sc_2 just beyond the fork of *Rs;* *Rs* very long, from two to two and one-half times R_{2+3}; *r* from one and one-half to twice its own length from the tip of R_1 and about twice its length beyond the fork of R_{2+3}; cell M_1 lacking; cell *1st* M_2 rectangular; *m-cu* about its own length beyond the fork of *M*.

Abdomen brown, with a more or less distinct median dark vitta on the tergites; pleural region darker brown. Male hypopygium relatively small. Ovipositor with the valves long and slender, acute, blackened basally, the tips paler.

Holotype, ♂, Vera Cruz, November 10, 1923 (A. M. Dampf). *Allotopotype*, ♀.

Paratopotypes, 6 ♂ ♀; ♂, Laboratorio Howard B. Cross, July, 1924.

E. pallidipes still further eliminates the characters available for separating *Penthoptera* from *Eriocera*.

Rhaphidolabis Osten Sacken.

Rhaphidolabis (Plectromyia) **tergata** sp. n.

Cell R_3 sessile; vein *M* very delicate, in alignment with M_{1+2}; *m-cu* shortly beyond the fork of *M;* male hypopygium with the tergite conspicuous, the lateral angles produced into ear-like lobes; mesal face of basistyle with a conspicuous pencil of long yellow setae.

Male. Length about 4.5 mm.; wing, 5.3 mm.

Rostrum and palpi black. Antennae relatively short, black throughout; flagellar segments oval. Head dark gray.

Mesonotum discolored; from slight patches of the normal coloration remaining, probably brownish gray, the praescutum presumably striped. Pleura brownish gray. Halteres short, brown, the base of the stem narrowly pale. Legs with the coxae light gray pruinose; trochanters brownish yellow; remainder of legs brown, the femoral bases paler, the terminal tarsal segments passing into dark brown. Wings (Fig. 11) with a pale brownish tinge; stigma vaguely delimited, slightly darker brown; an ill-defined brown cloud on the anterior cord; veins darker brown. Veins *M* and *1st A* very slender and pale. Venation: Sc_1 extending to beyond one-third the length of R_{2+3}, Sc_2 far before the origin of *Rs*, the distance being nearly one-half longer than that portion of Sc_1 beyond the origin of *Rs; Rs* gently arcuated; R_{2+3} elongate; R_2 about equal to $R_{1+}R_2$; R_3 a little shorter than R_{2+3}, gently arcuated; basal section of R_{4+5} very short, about equal to the basal deflection of R_5; *M* in alignment with M_{1+2}, both very pale, the latter becoming stronger beyond the fork of M_{3+4}; *m-cu* immediately beyond the fork of *M;* cell M_4 long and narrow, gently widened outwardly.

Abdomen dark brown, the hypopygium more yellowish brown. Male hypopygium (Fig. 19) with the ninth tergite (t) conspicuous, the caudo-lateral angles produced into ear-like lobes, the surface of the sclerite densely set with setiferous tubercles. Basistyle (b) produced into a slender lateral lobe that is spinous at apex, with a small sessile dorsal lobe that bears a lesser number of spinulae. Mesal face of basistyle with a conspicuous pencil of long yellow setae; slightly more basad of this, an oval cushion densely set with small erect setae. Dististyle (d) broad at base, the apex narrowed, the style extended beyond the apical lobe of the basistyle, the surface with a few large and numerous microscopic setulae. Interbases (i) slender, more or less foot-shaped. Aedeagus (a) broad.

Holotype, ♂, Ajusco, altitude 3500 meters, May 10, 1925 (A. M. Dampf); M. F. No. 549.

Tipula Linnaeus.

Tipula bellardiana sp. n.

General coloration gray; antennae with the basal flagellar segments bicolorous, black, with the tips paler; head with a narrow brown vitta; praescutum with three clearer gray stripes that are narrowly margined with brown, the median stripe further split by a capillary dark vitta; femora with a vague yellowish subterminal ring; wings variegated whitish subhyaline and brown, including a complete lunate band beyond the cord; tip of R_2 atrophied; abdomen obscure yellow, the subterminal segments blackened.

Male. Length, 10–11 mm.; wing, 13–14 mm.

Frontal prolongation of head brown, the nasus slender; palpi darker brown. Antennae relatively short, if bent backward extending about to the root of the haltere; scape and first flagellar segment yellow; intermediate flagellar segments more or less bicolorous, black, with the apex of the segment narrowly and sometimes indistinctly brownish yellow, more extensive on the basal segment, the amount becoming more restricted outwardly, the terminal flagellar segments uniform black; flagellar segments with the basal enlargement moderate in size, the verticils conspicuous. Head gray, the vertex with a median dark stripe.

Mesonotal praescutum yellowish gray with three clearer gray stripes, the median stripe further split medially and margined laterally with brown to form three approximated brown vittae; lateral stripes sometimes poorly indicated, usually appearing as a gray area margined with dark brown, the color crossing the suture onto the scutal lobes; scutal lobes gray, each with the antero-lateral marking just described and a much larger posterior spot that nearly covers the lobe; scutellum gray, the median area somewhat darker; postnotum dark brown, heavily pruinose. Pleura dark gray, indistinctly variegated with paler, this including the dorsal portion of the sternopleurite and most of the pteropleurite and meron; dorso-pleural membrane light yellow. Halteres obscure yellow, the knobs dark brown. Legs with the fore and middle coxae dark gray, the posterior coxae paler; trochanters reddish yellow; femora brown, the tips blackened, this color preceded by a vague obscure yellow ring of approximately equal width; tibiae light brown, gradually passing into brownish black, the tarsi concolorous; all tarsi longer than tibiae. Wings whitish subhyaline, variegated with brown, the stigma darkest; cell *C* infuscated except near the outer end where there is a clear spot; cell *Sc* yellow, alternating with four dark brown markings of slightly greater extent; wing-apex broadly darkened; a lunate brown mark before the cord, extending from the stigma to the wing-margin at Cu_1; an irregular pale brown cross-band across cells *R*, *M* amd *A*, near two-fifths the length of the former; bases of cells *R* and *M* darkened; the chief pale areas thus remaining include a curved band that completely crosses the wing beyond the cord, including the bases of cells R_2, R_3 and R_5 and almost all of cells *1st* M_2 and M_3; a Y-shaped band with the origin of *Rs* in the fork of the Y, the two distal pale blotches in cell *Sc* forming the arms; conspicuous pale areas in bases of cells *R* and *M* and including much of the Anal cells. Venation: Distal section of R_2 entirely atrophied; petiole of cell M_1 about one-half longer than *m*.

Basal abdominal tergites obscure yellow, the subterminal segments passing into dark brown, the segments with the lateral margin narrowly and indistinctly blackened, the mid-dorsal region with a black stripe that becomes broader behind; caudal margins of the segments narrowly ringed with pale; hypopygium reddish brown; sternites brownish yellow with an interrupted brown median vitta, the brown of the individual segments broadest behind, the color interrupted at the base of each segment. Male hypopygium with the ninth tergite large, reddish,

the caudal margin with a deep V-shaped notch, the lateral ears thus formed being very conspicuous, over-shadowing the dististyles. Basistyles very small. Ninth sternite with a U-shaped median notch. Eighth sternite unarmed.

Holotype, ♂, Chapingo, July 23, 1924 (A. M. Dampf); M. F. No. 254.

Paratopotypes, ♂, July 14, 1924, M.F. No. 235; ♂, July 21, 1924, M. F. No. 249; ♂, July 25, 1924, M. F. No. 255; ♂, August 8, 1924, M. F. No. 285; ♂, August 13, 1924, M. F. No. 291.

This interesting species of *Tipula* is named in honor of Professor Luigi Bellardi, pioneer student of the Diptera of Mexico.

Tipula oreomyzoides sp. n.

Antennae 16-segmented, elongate, the flagellar segments black, strongly incised beneath; general coloration of head and thorax gray, the praescutum with four darker gray stripes; wings tinged with brown, longitudinally vittate with whitish subhyaline, this color including cells *1st* M_2 and R_5; abdomen yellow, the tergites tri-vittate with brown, the sternites with a median interrupted brown stripe.

Male. Length, 12.5–14 mm.; wing, 15–18.5 mm.; antenna, 7.5–8 mm.

Frontal prolongation of head obscure yellow, the sides with a narrow dark brown line; nasus conspicuous; palpi dark brown. Antennae 16-segmented, elongate, if bent backward extending to opposite midlength of the abdomen; scape and basal portion of the first flagellar segment obscure yellow, the remainder of the organ black; flagellar segments deeply incised beneath. Head light gray, the center of the vertex a little darker gray.

Mesonotal praescutum light gray with four darker gray stripes, the narrower intermediate pair briefly connected at the cephalic end; scutal lobes gray, each with two darker gray spots; scutellum pale ashy gray; postnotum light gray. Pleura gray, vaguely variegated with darker, especially on the sternopleurite and meron; postnotal pleurotergite whitish gray; dorso-pleural membrane obscure yellow. Halteres pale, the knobs dark brown. Legs with the coxae obscure yellow, the fore coxae slightly pruinose; trochanters yellow; femora brown, paler at base, the tips passing into darker brown; tibiae brown, the tips broadly darker brown; tarsi brownish black; all tarsi longer than tibiae. Wings with a faint brownish tinge, the costal region more yellowish brown; disk of wings with a longitudinal vittate pattern of whitish subhyaline, this including the broad bases of the Anal cells; the narrow base and broader apex of cell *Cu;* bases of cells *R* and *M;* apical third of cell *M;* all of cells *1st* M_2 and R_5, and the base of cell M_3; longitudinal veins and the cord narrowly seamed with somewhat darker brown.

In a few cases, the wings are still paler than described, with the basal cells almost entirely whitish subhyaline; veins brown, paler in the costal region; obliterative areas including the cord and outer section of M_{3+4}. Venation: Distal section of R_2 pale but complete; cell *1st* M_2 irregularly pentagonal; *m* and petiole of cell M_1 subequal.

Abdomen yellow, the lateral margins of the tergites broadly brown, the line becoming wider behind, on the sixth to eighth segments including the caudal margins of the segments; an interrupted dorsomedian stripe, the vitta broadly broken at the posterior margin of each segment, less so at the base; hypopygium yellowish, the apices of the inner dististyles heavily blackened; sternites yellow, the caudal margins of the basal segments narrowly infuscated, on sternites six to eight more extensively so. Male hypopygium very similar in structure to that of *T. moctezumae* Alexander. Ninth tergite with the lateral lobes less conspicuously truncated at apices, the smaller median notch less abrupt. Eighth sternite with a small median liguliform lobe.

Holotype, ♂, Ajusco, altitude 3800 meters, in pine forest, May 10, 1925 (A. M. Dampf).

Paratopotypes, 2 ♂♂, altitude 3500–4100 meters, in pine forest, May 10, 1925. *Paratypes*, 7 ♂♂, Cerro de Hongos, altitude 3500–3700 meters, swept from grass in open pine forest, April 5, 1925 (A. M. Dampf); M. B. No. 117.

T. oreomyzoides is obviously closely allied to *T. moctezumae* Alex. and *T. azteca* Alex., which are likewise Mexican species from the higher mountains near Mexico City. The presence of sixteen antennal segments in *T. yreomyzoides* indicates that the range of antennal segments in *Tipula*, as now known, lies between 13 and 16. The supposed genus *Oreomyza* Pokorny is based upon three species from the higher Alps in which the males have 15-segmented antennae, the females 14-segmented antennae. The genus *Oreomyza* has no validity and must be placed in the synonymy of *Tipula*.

EXPLANATION OF PLATES.

Symbols: a—aedeagus; b—basistyle; d—dististyle; dd—dorsal dististyle; g—gonapophysis; i—interbasal process; p—phallosome; t—9th tergite; vd—ventral dististyle.

PLATE XI.

Fig. 1. Wing of *Limonia fissilis* sp. n.
Fig. 2. Wing of *Rhipidia* (*Rhipidia*) *multifida* sp. n.
Fig. 3. Wing of *Helius quadrifidus* sp. n.
Fig. 4. Wing of *Orimarga dampfi* sp. n.
Fig. 5. Wing of *Gonomyia* (*Gonomyia*) *remota* sp. n.
Fig. 6. Wing of *Teucholabis minuta* sp. n.
Fig. 7. Wing of *Pilaria rubella* sp. n.
Fig. 8. Wing of *Atarba mexicana* sp. n.
Fig. 9. Wing of *Atarba aperta* sp. n.
Fig. 10. Wing of *Eriocera pallidipes* sp. n.
Fig. 11. Wing of *Rhaphidolabis* (*Plectromyia*) *tergata* sp. n.

PLATE XII.

Fig. 12. Male hypopygium of *Limonia fissilis* sp. n.
Fig. 13. Male hypopygium of *Rhipida* (*Rhipida*) *multifida* sp. n.
Fig. 14. Male hypopygium of *Gonomyia* (*Lipophleps*) *strigilis* sp. n.
Fig. 15. Male hypopygium of *G.* (*L.*) *latistyla* sp. n.
Fig. 16. Male hypopygium of *G.* (*L.*) *spinistyla* sp. n.
Fig. 17. Male hypopygium of *G.* (*L.*) *bifida* sp. n.
Fig. 18. Male hypopygium of *G.* (*L.*) *haploa* sp. n.
Fig. 19. Male hypopygium of *Rhaphidolabis* (*Plectromyia*) *tergata* sp. n.

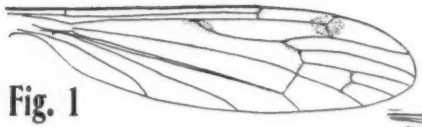

Fig. 1

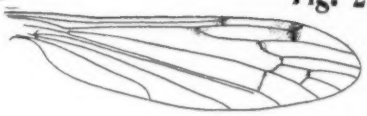

Fig. 2

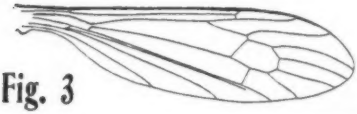

Fig. 3

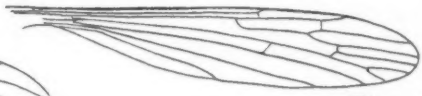

Fig. 4

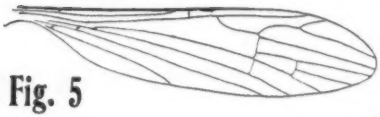

Fig. 5

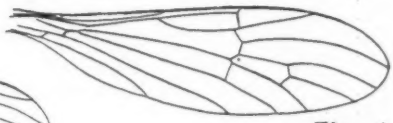

Fig. 6

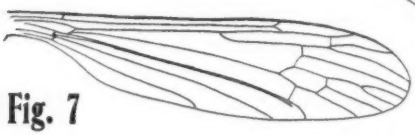

Fig. 7

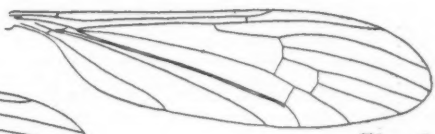

Fig. 8

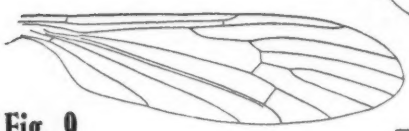

Fig. 9

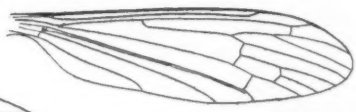

Fig. 10

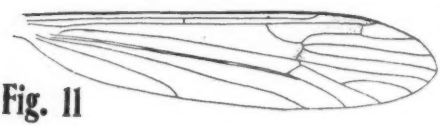

Fig. 11

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Fig. 12

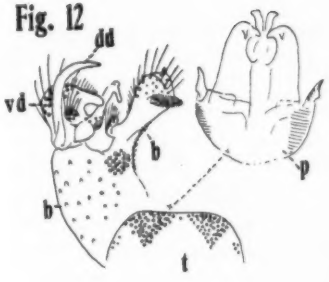
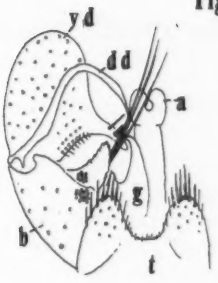


Fig. 13



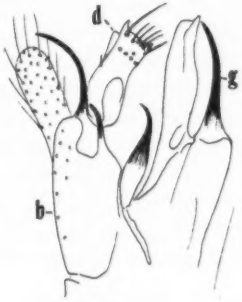


Fig. 15

Fig. 14

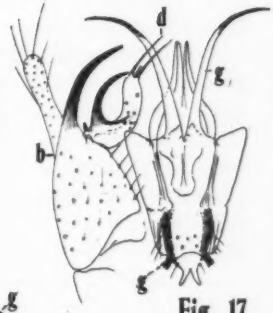


Fig. 17

Fig. 16

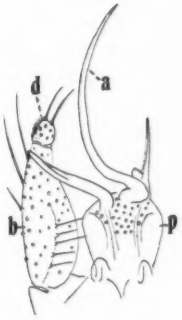


Fig. 18

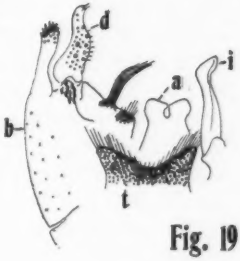


Fig. 19

C. P. Alexander

THE GREEN MUSCARDINE DISEASE IN SILKWORMS AND ITS CONTROL.*

R. W. GLASER, SC. D.

During the summer seasons of 1922, 1923, 1924 and 1925, a number of the writer's stock silkworms died of a fungus affection. The disease invariably appeared during hot, humid spells, and unless controlled assumed epidemic proportions. The eggs, from which these silkworm cultures were derived, originally came from healthy stock, and since the fungus has been described as a parasite of a variety of insects, it must have been introduced by one of them. When the affection first appeared, the writer assumed that he was dealing with the so-called green muscardine disease caused by *Metarrhizium anisopliae*. In 1924 dead worms and cultures were sent to Miss Vera Charles, mycologist in the Bureau of Plant Industry at Washington, who definitely identified the fungus as *Metarrhizium anisopliae* (Metsch.) Sorokin, and also gave much valuable advice. The fungus belongs to the family *Mucedineae*, one of the Fungi Imperfecti; those forms in which the ascigerous or perfect stage has not been found.

Metarrhizium anisopliae was first discovered by Metschnikoff (1) in 1879 as a parasite of the wheat cockchafer *Anisoplia austriaca*. The same year Sorokin seems to have studied the fungus. In 1880 Metschnikoff further found that the fungus was a parasite of the sugar beet curculio, *Cleonus punctiventris*. He says that the parasite produces an epizootic disease of the insects mentioned and believes it possible to use the disease economically to effect the destruction of pests by sowing the spores. In 1886 and 1888, Krassilstchik (2, 3) successfully used the fungus for combatting the sugar beet curculio. In 1895, Pettit (4) found the fungus parasitic on wireworms, *Agriotes mancus*, in New York State. Roherer (1910 and 1913) (5, 6), in Trinidad, found froghoppers, *Tomaspis postica*, to be extremely susceptible and described favorable field experiments with the fungus. Speare (1912) (7, 8) investigated

*From the Department of Animal Pathology of The Rockefeller Institute for Medical Research, Princeton, N. J.

the same fungus in Hawaii. Here the sugarcane borer beetle, *Rhabdocnemis obscura*, is affected. Speare, from his own experiments and reports by other investigators, concludes that it is quite evident that the fungus is more virile for some hosts than for others. Stevenson (1916) (9), in Porto Rico, attempted to combat white grubs, which are serious pests of sugar cane, with the parasite. Infection experiments performed in cages were successful, but field trials failed.

Two forms of *Metarrhizium anisopliae* have been discussed, by Pettit (4) in 1895, and by Johnston (10) in 1915. Johnston is of the opinion that the two forms simply represent physiological races of the same fungus, *i. e.*, that the morphological and other changes described do not warrant the assumption that two distinct varieties exist, but that these changes are dependent upon the host or upon the artificial medium.

Among useful insects, the fungus has so far only been described from the silkworm. It was discovered on this form in France in 1893 and was studied by Delacroix (11) who named it *Oospora destructor*. No recent work on the parasitism of *Metarrhizium anisopliae* exists, especially, nothing dealing with the disease produced in silkworms. For this reason, the writer undertook a study of the subject.

Infection Experiments.

The spontaneous cases in silkworms, when in the early stages, are difficult to recognize. The later stages are easily distinguished, because they are usually accompanied by a yellowish skin tint and after death by rigidity of the body followed by mummification. After death, the body soon becomes covered with a white hyphal down which turns green as the spores develop in a day or two more. However, for a study of the progress of the disease, experimental infections must be produced. From such infected worms blood films were prepared daily and studied both in the fresh condition and in the fixed and stained state. A worm or two from each experiment was daily saved for histologic study.

Previous workers have emphasized the importance of a humid, warm environment for the development of green muscardine. The same condition seems necessary in the case of the silkworm, for spontaneous cases of the disease were rarely found in the stock excepting during humid, hot weather. If

such weather persisted any length of time, extermination of a large part of the stock always seemed inevitable until certain control measures were instituted.

To produce a humid environment, the infected worms were placed in battery jars and fed daily with mulberry leaves slightly moistened with water. Pieces of glass with a small hole or two bored through, to allow enough fresh air to enter, covered the battery jars. In this manner a slightly moist atmosphere can be maintained as evidenced by the continual cloudy appearance of the glass container. Too much moisture, such as might be produced by inserting a saturated piece of sponge or cotton, breeds other troubles. It is not often necessary to produce artificial warmth during June, July, August, and September in the region (New Jersey) where these experiments were conducted.

An attempt was first made to infect healthy silkworms by feeding them with a water suspension of the green *Metarrhizium* spores obtained both from cultures and from cadavers. Fourth or fifth stage silkworms can easily be fed such a suspension if they are kept away from all moisture and food for from 12 to 24 hours. After this time, if held ventral side up and restrained in one hand by holding the head, they will imbibe two, three, or more small drops of liquid, carefully placed over their mandibles with a fine capillary pipette. Care must be taken not to contaminate the silkworm skin with the spores, otherwise it will be difficult to tell how infection occurred. One dozen worms were fed heavy suspensions of ripe spores of the fungus obtained from cadavers, and one dozen were fed with spores from cultures in the manner described, but in no case was the disease reproduced. Apparently then, worms do not acquire green muscardine by swallowing the spores.

Two dozen silkworms of the 4th and the 5th stage were then infected by painting the skin in places with moistened camel's hair brushes dipped in the spores from cadavers and from cultures on potato. The mortality from muscardine in this experiment was 100 per cent, the animals dying either in the larval or early pupal stage. Fig. 1, Plate XIV, shows a group of the worms dead of green muscardine. The darker worms are entirely covered with olive green spores which may be removed in flakes. The white areas shown on two of the worms represent places where the hyphae have as yet not fruited.

Fig. 2, Plate XIV, shows some early pupae that died of the disease and were removed from their cocoons. On these the fungus has not fruited and is consequently white. It is rare for the fungus to fruit on mature larvae, and on early pupae after they are enclosed within the silk. The lack of the necessary moisture, air, or both, within the dense cocoon is probably responsible for this condition.

The course of events, observed from infection to death and after, was traced from further infections. The spores germinate on the skin of the silkworms and soon penetrate the body. If some blood is removed from the infected worms in two or three days, a picture represented in Group 1-6, Plate XIII, will be seen. Figures 1, 2, 3, and 4 represent growing fungus bodies; Figs. 5 and 6 two blood cells. Fig. 3, Plate XIV, shows a section through a worm in this condition. Above some muscle tissue is seen, whereas below in one of the cavity spaces large numbers of fungus bodies and blood cells are found. A reaction on the part of the host occurs, indicated by large masses of phagocytes which congregate around and between the fungus. The phagocytes are, however, unable to cope with the parasite which is too large for ingestion. Worms when in this state never recover, and it is doubtful if they are ever able to gain control of the parasite once it has gained access to the body cavity.

On examining the blood in about 4 or 5 days after infection, one finds besides such stages as represented in Figs. 7 and 8, Plate XIII, also larger forms as shown in Fig. 9, and the beginning of branching as illustrated in Fig. 10. At this time the worms begin to lose their appetite, assume a yellowish color, and appear sick. During life the cells of all the tissues are able to resist the invasion of the parasite. The parasites are found only in the body cavity spaces, *i. e.*, in the blood plasma. In 5 or 6 days, the worms become extremely feeble and usually die. After death they become rigid (in contradistinction to the flaccid postmortem condition produced in flacherie or in jaundice) and brownish yellow in color. The body fluids when examined after death reveal such forms as represented by Figs. 11, 12, 13, 14, and 15, Plate XIII. Mycelium formation with the occurrence of branching has become extensive. The mycelia are segmented. During this period invasion of nearly all the body tissues occurs accompanied

by disintegrative changes of the tissues. Fig. 4, Plate XIV, demonstrates one of the longitudinal muscles invaded by the parasites. The strands or bundles of fibrillae are shown to have separated. In a day or two more the mycelia ramify throughout the body causing the cadaver to become extremely rigid and somewhat hard and brittle. At this time there is also a tendency for certain of the mycelia to penetrate outwards. Fig. 5, Plate XV, shows such a hyphal thread penetrating through the hypodermis. In 7 or 8 days after artificial infection the hyphae break through the skin everywhere and the worms become entirely covered by a white down. This white down consists of the hyphae destined shortly to become the sporophores. After a day or so more, the rather short and branched sporophores begin to form spores, as shown in Fig. 18, Plate XIII.

The spores soon cover the entire body of the silkworms, and as they mature in a day more become olive green. The mature sporophores cohere, and for this reason large flakes may be removed with a knife blade or forceps. The spores are produced in succession on the sporophores and also cohere until disturbed. They vary from 5-7μ long to 3μ wide and are oblong, as shown in Fig. 16, Plate XIII. The spores are enclosed in a resistant membrane. The writer has kept spores in a dry condition for three years and finds that after that time they are still capable of germination. They are also quite resistant to heat and disinfecting agents. Germination does not seem to be affected by heating a suspension of spores in water for one hour at 60°C. Dry heat of 180°C., applied for one-half hour, kills them. They are killed when autoclaved for 20 minutes or boiled in water for 10 minutes. Short exposures to 5 per cent carbolic acid or to 1:1000 corrosive sublimate solution have no effect. Five per cent carbolic acid, applied for from 3 to 6 hours, and corrosive sublimate 1:2000, applied for one-half hour, are effective.

As previously stated, the host usually dies in the larval stage. However, sometimes, especially among those infected during the last instar, some individuals will spin a cocoon and succumb within mummifying later. In such cases the white hyphal down appears, but the fungus rarely fruits.

From the results presented in this study of the forms of the fungus appearing in silkworms, no evidence for the existence

of a perfect stage was obtained. The conidial stage develops so readily in the worms and on artificial culture media that a perfect stage is apparently unnecessary for the continuance of the species and none may exist.*

Development on Media.

Previous workers have given such a good account of the development of *Metarrhizium anisopliae* on artificial media that an account here would constitute merely a repetition. Suffice it to say, that the fungus grows very readily on potato and potato agar. Germination, as observed in hanging drops of the spores, begins in a few hours and is completed in about a day. The thick spores swell somewhat and throw out germ tubes frequently at both ends. Fig. 17, Plate XIII. After this the tubes elongate and form segmented mycelia as shown in Fig. 6, Plate XV. These form a white down over the surface of the media. The short dividing and thick stages found in silkworms were not found on media. After a few days the olive green spores are again formed on the sporophores which push up. On plates, colonies develop a rounded stellate form, Fig. 7, Plate XV, and show dark fruiting and light non-fruiting areas.

To obtain the development of the fungus on media, one must start with spores. Media inoculated with the non-fruiting forms from larval blood always remained sterile. This is curious in view of the fact that fungus development will progress normally when these forms found in diseased cases are injected into the body cavity of normal worms by means of a syringe.

Control Measures.

The *Metarrhizium* spores, as shown, are quite resistant, and since they are produced in enormous numbers, which are easily distributed by air currents, silkworms or other insects, etc., it is necessary to keep the fungus from fruiting and dis-

*From the description of the various stages of the parasite found in silkworms it must not be supposed that these correspond in every case with the time given. At times a certain extent of overlapping of forms occurs within one period. In general, however, after a study of many worms in all stages of the disease, the writer feels that a rather definite or circumscribed series of stages correspond with a more or less definite time expressed in hours or days. Variations, as stated, are found depending on temperature, humidity, and possibly other factors.

tributing itself, once it appears in a silkworm culture. This is not as easily accomplished as one would at first suppose. To clean or sterilize everything, excepting the worms and their food (which cannot be sterilized), is a foregone conclusion. To remove and destroy all diseased worms as soon as the white hyphae appear and before the parasite has sporulated is also readily done. However, exceptional cases are those that often breed all the trouble.

At times, especially before moulting when the worms do not eat and when the old skin becomes naturally somewhat discolored, it is difficult superficially to decide whether fungus exists or not. If fungus bodies should be present within such doubtful worms, and if the weather at the time should be hot and humid, the hyphae might penetrate the skin and fruit during the night. In such cases distribution of spores by air currents and especially by other silkworms, followed by mass infection, will occur before the infected worm can be removed and the container sterilized. This has actually occurred twice, in certain broods of the writer's silkworm cultures, during the past four years. The only efficacious method used, when fungus disease was suspected in a brood, consisted in an examination of the blood of all the worms in this particular brood. If fungus bodies were found in certain worms, these were immediately killed and burned. Those worms that showed negative findings were held for three or four more days, to permit the infection to advance if present, and were then subjected to another blood examination. A positive finding again resulted in the destruction of infected worms; a negative one was considered evidence of non-infection. This method has proven practical and has permitted us to keep the disease under control until all green muscardine was eliminated. When worms are properly handled and not overcrowded green muscardine appears only sporadically in certain broods or cultures and can be readily controlled as outlined.

The regulation of humidity is another very important factor when green muscardine exists. During the dangerous period the jars, trays, or other containers in which the worms are reared, should be kept dry. The containers must be left open during this time, and the food must not be tendered while in a moist condition; mulberry leaves wet from washing, rain or dew should be dried first, not withered. The rearing

room also must not be washed and the atmospheric moisture partially controlled by opening or closing the windows, dependent upon the outside weather, and by using an oil heater for a short period when the barometric pressure increases appreciably.

Summary.

Green muscardine, a fungus disease caused by *Metarrhizium anisopliae*, appeared among some of the writer's silkworms. From a review of the literature, it is shown that green muscardine affects many insects of economic importance. Reports on its use in combatting noxious insects are conflicting. This may be dependent on differences in susceptibility among the different species, or on climatic differences, or on differences in the methods used. Among beneficial insects, the silkworm is the only form in which green muscardine has been found.

Successful infection experiments are described. Silk-worms do not acquire the disease by swallowing the spores, but when the spores lodge on the skin germination and infection soon occur. The symptoms of diseased worms and the postmortem picture are described. The time and course of events within the worms from infection to death and after; the various stages of the fungus found and their pathological effects on certain tissues are described. The examination of blood is a reliable and useful index of the condition of the worms. No evidence for the existence of a perfect stage of *Metarrhizium* was obtained.

The best media for the cultivation of *Metarrhizium anisopliae* is mentioned and its development in culture outlined.

Control methods which have proven practical are described. These are primarily based on blood examinations and on humidity regulation. It is shown that if the fungus can be kept from sporulating its spread can be prevented.

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EXPLANATION OF PLATES.

PLATE XIII.

Sketches

Figs. 1–6. Group of fungus bodies and blood cells found in blood of silkworms two or three days after experimental infection of the skin with *Metarrhizium* spores.

Figs. 7–10. Fungus stages found in blood of silkworms four or five days after infection.

Figs. 11–15. Fungus stages found in silkworm body fluids after death.

Fig. 16. Ripe fungus spores.

Fig. 17. Germinating fungus spore.

Fig. 18. Sporophores of *Metarrhizium* bearing ripe spores.

PLATE XIV.

Photographs. Natural Size.

Fig. 1. Group of silkworms dead from green muscardine.

Fig. 2. Group of early pupae removed from cocoons and dead from green muscardine.

Photomicrographs. ×810.

Fig. 3. Section through a worm three days after experimental infection of skin with *Metarrhizium* spores. Above normal muscle tissue. Below a body cavity space containing large numbers of fungus bodies and blood cells.

Fig. 4. Section through a worm which died six days after experimental infection. One of the longitudinal muscles invaded by the fungus.

PLATE XV.

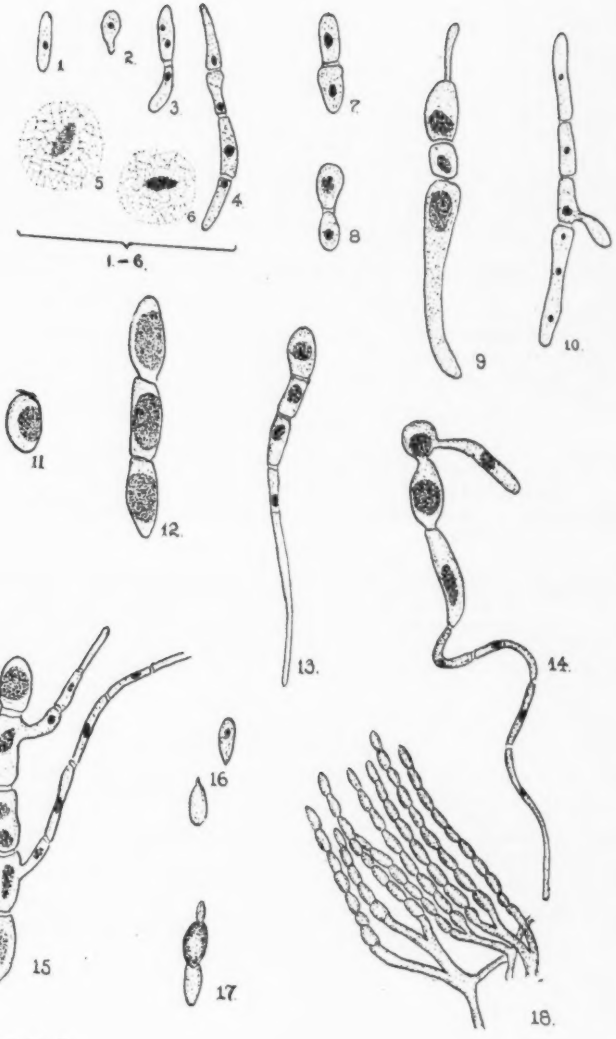
Photomicrographs. × 810.

Fig. 5. Section through a worm which died seven days after experimental infection. Hyphal thread is seen penetrating through the hypodermis of host.

Fig. 6. *Metarrhizium*, from a five day old potato culture, showing the segmented mycelia.

Photograph. Natural Size.

Fig. 7. Colonies of *Metarrhizium*, on potato agar plate culture eight days old, showing rounded stellate form with fruiting and non-fruiting areas.



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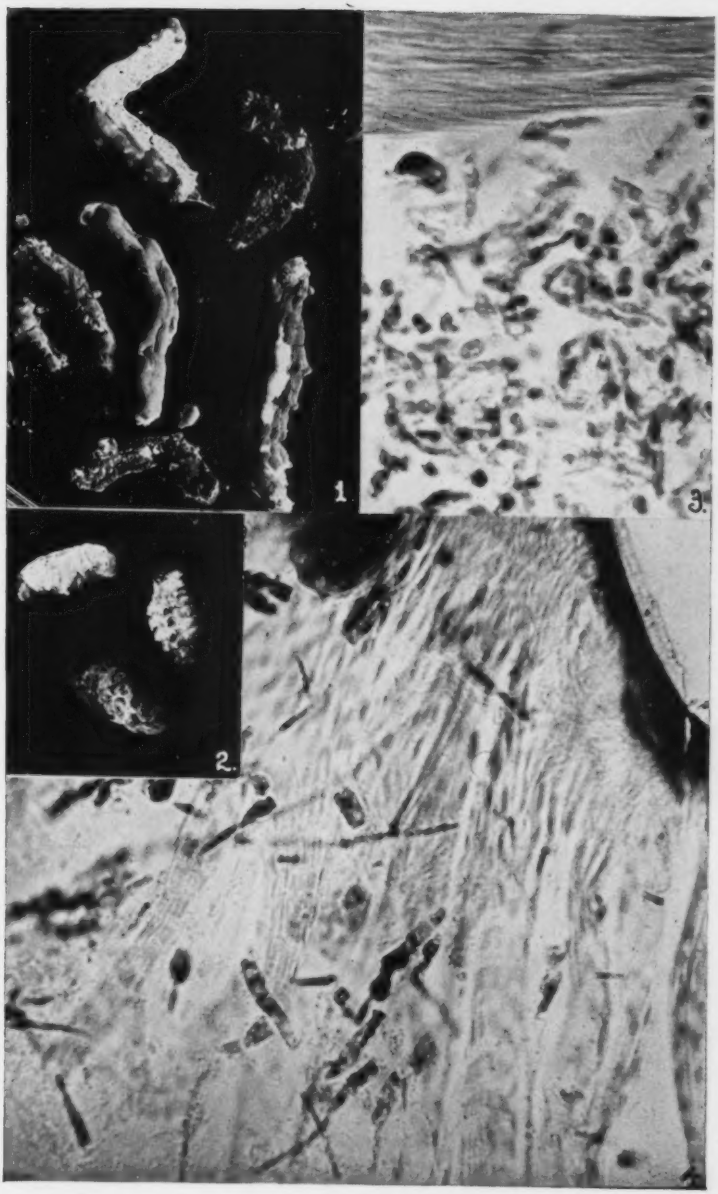

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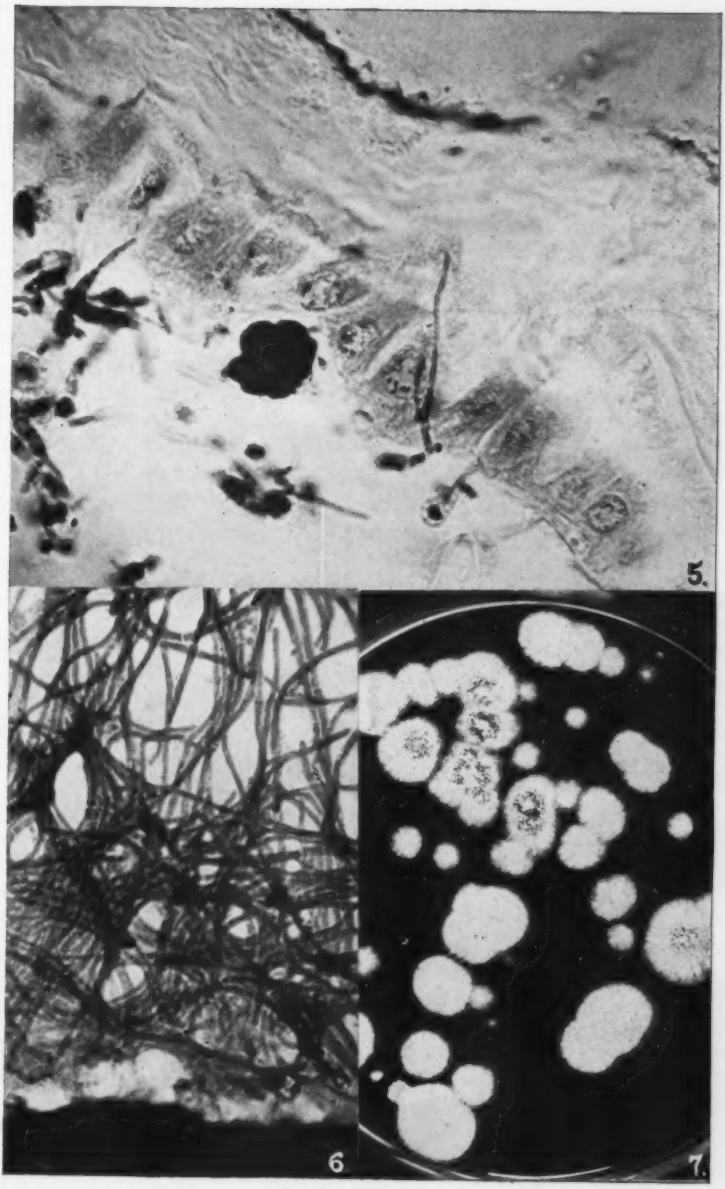

R. W. Glaser, Sc. D.

FURTHER EXPERIMENTS ON A BACTERIAL DISEASE OF ADULT FLIES WITH A REVISION OF THE ETIOLOGICAL AGENT.*

R. W. Glaser, Sc. D.,

In 1924[1] the writer recorded a bacterial disease of adult house flies caused by a *staphylococcus* which was described under the specific name of *muscae*. The disease was recognized by the fact that certain flies reared in captivity, and approximately two weeks after emergence, developed swollen and translucent abdomens which could not be correlated with feeding nor with the development of the ovaries or testes. It was found that this distension was due to a clear, sticky, serous fluid devoid of cellular blood elements, but containing large numbers of a *staphylococcus* which was easily cultivated. Certain other external appearances, such as a paralysis of the legs, were frequently present. The internal cytolizing effects of the bacterium upon the tissues with the probable inhibition of the functions of the Malphighian tubes, leading to an edematous condition, was outlined. It was found that the disease is sporadic in its appearance and never assumes the form of an epidemic. About 50 per cent of adult flies contract the malady when experimentally infected by permitting them to feed on the abdominal fluids of diseased individuals, or on bouillon or other cultures of the microorganism. Experimentally infected flies die in from 7 to 29 days with a mean of 16+ days. Symptoms appear in from 2 to 5 days prior to death.

In 1924, it was felt that not much may be expected from the disease economically, *i. e.*, as an adjunct in controlling house flies. This assumption was based on several facts of which the principal ones appeared to be: (1) Males were found to be more than twice as susceptible as females. (2) The disease is rather long in developing; it is, strictly speaking, a disease of senility and in consequence most of the females oviposite before showing symptoms. (3) The causative microorganism is of rather low virulence compared with many other bacteria

*From the Department of Animal Pathology of The Rockefeller Institute for Medical Research, Princeton, N. J.

pathogenic to insects. (4) It was found that temperature also has an important relation to the disease. Flies acquire the infection much more readily during hot weather than when the weather is cool.

From the foregoing it seemed, therefore, advisable to attempt to increase the virulence of the staphylococcus, preferably by successive passages through flies. A large series of experiments were made with newly emerged flies whose feces had first been "tested out" and found free of the microorganism in question. The bacterium was successfully passed through 3, 4, and 6 separate lots of adult house flies from broods of the same generation without the intervention of cultures and without waiting for disease symptoms to develop. In this set of experiments, the feces and vomit spots of infected flies were dissolved in nutrient bouillon and fed to the following series. At the same time plates were poured, and streaked with some of this material and incubated to make sure of the continued existence of the staphylococcus within the alimentary tract. After 24 to 48 hours any characteristic colonies which developed were picked, isolated, and later studied using the principal diagnostic characters appended at the end of this article. After infection the microorganism, with few exceptions, became established in the alimentary tract. Twenty-four hours was the time consumed in each series between infection and recovery of the infected feces, so that in the case of the six passages approximately twelve days were consumed.

Next the bacterium was successfully passed through six different generations of adult flies. In this case the writer waited until the disease developed in a certain number. Since this takes a long time, as previously noted, separate generations of flies must be used for the passages, since the life span of the average fly is not sufficient and the bacterium must be recovered in cultures between successive passages. The staphylococcus was recovered in cultures from the abdominal fluid, and again fed to a fresh generation of adults, which were derived from broods fed and mated so that the flies would emerge within a few days of the time when needed. This was done so that the staphylococcus would not remain on the medium long, and again lose any virulence that might have been acquired in passage. Control flies fed with sterile food accompanied all infections. No disease appeared among these, nor was

the staphylococcus recovered on agar plates prepared from feces.

On analyzing all of these data, the writer finds that the virulence of the staphylococcus was not enhanced through passages. Approximately between 40 and 50 per cent of the infected adult flies contracted the disease, no matter whether the microorganism was derived from fresh spontaneous cases, from cultures kept alive for 2 years by transplanting on agar, or from any of the experimental passages. It was also noticed that the mortality was higher during hot days (80°–90°F.) than during cooler days. These observations are interesting and agree with Paillot's experiments[2] performed on a number of different species of insects in 1922. Paillot also found that the virulence of bacteria, pathogenic for the insects investigated, could not be enhanced through passages. Differences in pathogenicity exist, but these are either positive or negative quantities to which or from which one cannot add or subtract as the case may be.

It was next thought that if the infection could be started during the larval stage, it might possibly manifest itself earlier in adult life and thus prevent the female flies from laying their eggs. A large crop of newly emerged adults was divided into two lots, containing each the same number of males and females, and each lot placed in a separate breeding jar containing fresh horse manure.[3] Both jars were fed alike; the adults received sugar and sterile nutrient bouillon for approximately two weeks and until most of the females had oviposited. The manure in one jar had been contaminated with a 24 hour bouillon culture of *Staphylococcus muscae* prior to the introduction of the adult flies. The other jar constituted a control. Both were carefully examined from time to time. No dead larvae were found. As soon as the next generation of adults emerged, the same number were extracted from the experimental as from the control jar and segregated in sterile, 6 ounce, wide mouthed bottles covered with cheesecloth. While in these bottles the flies were fed and observed daily until the conclusion of the experiment.

No flies died of disease in the control bottles. In the experimental bottles, after the prescribed time and after most of the females had oviposited, approximately one-fifth of the flies contracted the disease. Again, the greatest number of

deaths were obtained during spells when the temperature was rather high. It, therefore, seems impossible to shorten the time from infection to death by contaminating the larval environment.

During the course of this work many pure cultures of the staphylococcus were examined and compared with original stock cultures isolated in 1923. While engaged in these examinations a number of minor, but useful, new diagnostic characters appeared which had been previously overlooked. Since many species and varieties of staphylococci have been described, and since superficially the form in question resembles *S. pyogenes aureus* and *albus*, it was thought advisable to redescribe *S. muscae* in order to emphasize its divergent characters. One or two minor errors in the original description are also corrected.

Staphylococcus muscæ Glaser (1924).

1. *Morphology:* Gram-positive staphylococcus. 24-hour bouillon culture, chains of 5 units common; 24-hour agar slant, single or double forms. Diameter 0.5-1μ. At room temperature about twice size of forms at incubator temperature; 37°C., non-motile. Capsules absent.

2. *Cultural characters: Nutrient bouillon*, 24 hours, 37°C., clouding strong, no ring, sediment, viscid on agitation; no odor. 48 hours, ring. 96 hours, clear, pellicle, sediment abundant and extremely viscid on agitation. 24 hours, room temperature, clouding faint with flocculent masses; ring, pellicle, sediment abundant and viscid on agitation; no odor. 48 hours, same. 96 hours, same with clearing. *Agar stroke:* 24 hours, 37°C., white, abundant, echinulate with rounded margins, flat, glistening, smooth, opaque, odor absent, consistency viscid. 48 hours, same. 96 hours, same. In 2 weeks, still white, shiny and smooth. 24 hours, room temperature, white, moderate, echinulate with rounded margins, dull, rough, opague, odor absent, consistency butyrous. 48 hours, abundant, dry, rough and wrinkled. 96 hours, cream colored to light yellow, very much wrinkled, otherwise same. In 2 weeks, cream colored, or 3 chroma of orange yellow according to Winslow's color chart.[4]

Potato: 24 hours, 37°C. Abundant, cream colored.

Agar colonies: 24 hours, 37°C. Growth rapid; white; form round; surface smooth; colony flat; internal structure, amorphous; diameter 0.4–4.5 mm. 48 hours, form irregularly round; white; shiny; surface smooth with faint radiations from center to periphery; elevation, flat with center of colony slightly raised as a cone. 96 hours, same. 24 hours, room temperature, growth moderate; white; dull; form irregularly round; surface rough; elevation flat; internal structure amorphous. 48 hours, colonies well developed; surface rough with pro-

nounced radiations near periphery; elevation flat with center of colony; slightly raised as a cone. 96 hours, cream to light yellow; very much wrinkled; radiations from center more distinct.

Gelatin colonies: 48 hours, 22°C., growth moderate; small colonies; dull white; rough; no liquefaction. 96 hours, growth good; small colonies; rough; form irregularly round; elevation from center of colony edge undulate; liquefaction saucer; internal structure coarsely granular.

3. ***Biological reactions:*** *Milk:* 48 hours, 37°C., not coagulated. 96 hours, coagulated. Extrusion of whey. Reaction in 1 week from pH 6.6 to pH 5.5. Reaction of whey in 2 weeks pH 5.4. Rennet tests negative. *Gelatin stab:* 24 hours, 20°C., growth, best at top; line of puncture beaded; liquefaction infundibuliform, complete in 6 weeks although not all of gelatin is affected. *Horse blood agar:* 24 and 48 hours, 37°C., no hemolysis. *Relation to oxygen:* Aerobe. *Indol production:* Absent. *Pathogenicity:* Pathogenic for adult house flies (*Musca domestica* L.).

Table I gives the maximum acidity reached in 72 hours at 37°C., as well as the change in acidity produced at that time by the staphylococcus grown in bouillon containing 1 per cent of various carbohydrates. The lack of gas production is also recorded.

TABLE I.

Carbohydrate used	pH of bouillon	pH produced in 72 hours	Change in pH produced in 72 hours	Gas
Glucose	7.4	7.0	−0.4	—
Lactose	7.2	6.8	−0.4	—
Sucrose	7.3	6.8	−0.5	—
Maltose	7.4	7.7	+0.3	—
Mannite	7.6	7.3	−0.3	—

Summary

The virulence of *Staphylococcus muscae* was not enhanced by successive passages through adult house flies. Although flies in the larval stage do not develop the disease described, it is possible to infect them by contaminating their environment with the microorganism. The disease. develops subsequently in a certain number of the adults. It is, however, impossible by this procedure to shorten the period from infection to death. Of external contributing factors temperature, and of internal contributing factors age and sex are important, but these do not appear sufficient. Resistance or

susceptibility seems to depend on other unknown causes within the body organization of the insects. A revision of the diagnostic characters of *Staphylococcus muscae* is appended.

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CERTAIN GENITALIC CHARACTERS IN LASPEYRESIA MOLESTA BUSCK., AND LASPEYRESIA PRUNIVORA WALSH.*

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During the past summer the writer was confronted with the necessity of separating the adults of *Laspeyresia molesta* Busck., from *Laspeyresia prunivora* Walsh. While the two species are very easy to separate, when in good condition, on the characters of the coloration of the scales of the wings, it was found difficult, at first, to recognize material rescued from bait-pails used to trap these moths. In making weekly collections from such bait-pails, the moths were frequently submerged, denuded and in what seemed a hopeless condition for determination. After a study of the genitalia of the males and females of these species, as well as *L. interstinctana* Clem., a closely related species, tangible characters were discovered by means of which the species could be readily and quickly determined.

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In general the presence of the spurious vein in the forewing, the absence of the stalking of Rs and M_1 in the hind wing, the short proboscis, the shape of the palpi with a small terminal segment, and the position and number of the ocelli, helped to separate the species of Lasperyesia from other Tortricids found in the bait-pails.

The number of species of moths taken from bait-pails was considerable, but as far as the writer is aware, only three species of Laspeyresia were attracted to the pails. *L. molesta* Busck., was found in large numbers, sometimes as many as 106 in a single pail, *L. prunivora* Walsh., was much less numerous, while *L. interstinctana* Clem., was taken but rarely. The comparative abundance of these species helped greatly in separating them.

The genitalia of the three species of Laspyresia are similar but present differences sufficient to lend specific characters. No attempt was made to describe the genitalia in detail, but merely to point out vivid characters of value to an economic entomologist working under the difficulties experienced by the writer.

Certain secondary structures, no doubt the "Haartschen" of Poljanec (1901)* or the "Anal brush" of Newell (1918),† were found more valuable in the males than the genitalia proper. In *molesta* they are filamentous, elongate, and appear as two delicate feathers, one on each side of the anus attached to the conjunctiva between the eighth and ninth segments. In *prunivora* they are short, flattened and plate-like, while in interstinctana they are similar to those in *molesta*.

The shape of the harpes was also found valuable for separating the males of these species. In *molesta* the cucullus is large and about the same size as the sacculus, or but slightly smaller. The size of the two portions of the harpes is due to the neck which occurs nearly in the middle of the harpes, and is greatly constricted. In *prunivora*, the neck is not conspicuously constricted, occurring closer to the cucullus than to the sacculus, resulting in a very small cucullus and a comparatively large sacculus. In *interstinctana*, the cucullus is distinctly larger than the sacculus, and the neck or constriction, as in *molesta*, is prominent.

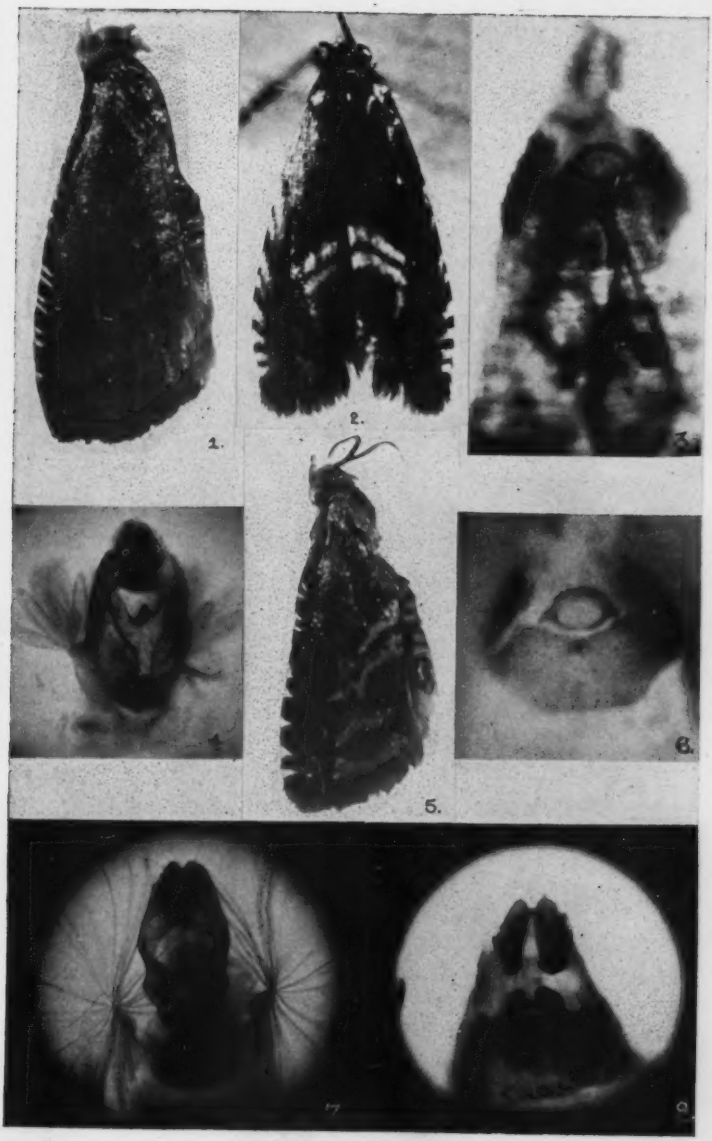
*Poljanec, L Arb. Inst. Wien, 13: 155-196, 1901.
†Newell, A. G. Ann. Ent. Soc. Am. 11:109–156, 1918.

The characters in the female are likewise distinctive. In *molesta* the chitinized plate surrounding the opening of the vagina is produced into a spine-like point at each posterior edge. There are also two finger-like processes adjacent to the vaginal opening. In *prunivora* the chitinized plate is somewhat trapezoidal in shape without the spine-like projections and without the finger-like processes adjacent to the vaginal opening. The female of *interstinctana* was not examined.

No doubt further studies of the uncus, vinculum, gnathos, and socii will reveal additional characters of value but for practical purposes in separating these species, the characters mentioned will be found important.

EXPLANATION OF PLATE XVI.

Fig. 1. *Laspeyresia molesta* Busck.
Fig. 2. *Laspeyresia interstinctana* Clem.
Fig. 3. Tip of the abdomen of *L. prunivora*, showing vaginal opening and Bursa copulatrix within the body.
Fig. 4. Male genitalia of *L. prunivora* Walsh.
Fig. 5. *Laspeyresia prunivora* Walsh.
Fig. 6. Vaginal opening and plate of *L. prunivora*.
Fig. 7. Male genitalia of *L. molesta* Busck.
Fig. 8. Female genitalia of *L. molesta* Busck.



S. W. Frost.

CONTRIBUTION TO THE KNOWLEDGE OF THE INTER-RELATIONS OF THE BUMBLEBEES OF ILLINOIS WITH THEIR ANIMATE ENVIRONMENT.*

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INTRODUCTION

Tho bumblebees are well endowed by nature with the means of a certain type of defense, this does not make them invulnerable to the attacks of parasites nor secure to their sedentary nests an immunity from inquilines, scavengers, and the like. Even adult bumblebees occasionally fall victims to predaceous insects and other animals, in spite of the fact that they possess a formidable sting and know how to use it to very good advantage.

It is important to note that among the motley collection of insects which I have found in the nests of bumblebees or which have been recorded as being found in such nests, there is not a single species which is known to enter into trophallactic relations with its host. This is in general keeping with the lack of trophallaxis between adults and immature forms of bumblebees. Accordingly, it can not be said that at the present time trophallaxis plays an important part in the contact of bumblebees with their animate environment within the nest. Of course, the possibility of a trophallactic relation between the young and adult bumblebees at a past time cannot be altogether disregarded. In contrast, it is to be noted that much evidence is now available to point "to trophallaxis as the source of the social habit in wasps, ants and termites" as stated by Wheeler (1918). As already intimated, the interrelations of bumblebees and other insects is all to the advantage of the latter, and tho often not detrimental to the bumblebees it may be decidedly so.

*Contribution from the Entomological Laboratories of the University of Illinois, No. 100. Extract from a thesis presented to the Faculty of the Graduate School of the University of Illinois in May, 1923, in partial fulfillment of the requirements for the degree of doctor of philosophy.

Outside of the nest, however, we are presented with a most remarkable interrelation between bees and flowers, a relationship which is beneficial to both the bumblebees and flowers. Space is too limited to permit a consideration of the wonderful interrelation existing between flowers and bumblebees. It suffices here to point out that all Illinois species of bumblebees are polytropic and there is every indication and reason to believe that all of the other American species are polytropic. In fact, it is doubtful whether there exists an oligotropic species of bumblebee anywhere in the world, tho one species in Europe was once reputed as such. The long time required for the development of the immature stages, the progressive feeding, and the social life of bumblebees prohibits their dependence upon a single species of flowering plant. The structural adaptations of both flowers and bees to their intimate relations is truly a wonderful thing and "if it were not so common or if it existed only in the South Sea Islands would be one of the wonders of the age" (Z. P. Metcalf). It is because of this interrelation between bumblebees and flowers that bumblebees are beneficial insects, and chiefly because of the part that they play in the fertilization of the red clover are classed among the economic insects. In order to produce red clover seed in New Zealand it was necessary to import bumblebees to that country from England. The introduced species have now become thoroly established and red clover seed is now produced (Dunning, 1884, 1886; Smith, 1896; Graenicher, 1910; Thomson, 1922).

It is the intention of the writer to present in this paper a brief account of the enemies and inquilines which, directly or indirectly, affect the existence of the bumblebees in the region centering about Illinois. A detailed description of certain stages of the more important parasites, whose status is briefly outlined at this time, will be left for a later paper.

My thanks are due to several persons for their kindness in naming material and acknowledgment is made in the text for these determinations. The writer is particularly indebted to Mr. C. F. W. Meusebeck for furnishing the redescription of *Apanteles nephoptericis* (Packard), and the redescription is given here for reasons stated further on in the course of this article.

For the sake of convenience my remarks have been grouped under several headings.

A. Social Parasites

Much has been written about the habits of the social parasites of bumblebees, belonging to the genus *Psithyrus* of the family Bremidae (Bombidae.) Until a very recent date, however, this literature dealt only with the European species. The first real contribution towards a settlement of the true status of *Psithyrus* in the New World—not speculation—was the short paper by Sladen in 1915. Since then the writer (1916 and 1921a) has published two short articles concerning *Psithyrus* in the nests of bumblebees, and more recently Plath (1922a and 1925) has published accounts of his extremely interesting and valuable observations. The following observations, comments and résumé of the literature concerning the only two species known to occur in Illinois, *Psithyrus variabilis* (Cresson) and *Psithyrus laboriosus* (Fabricus), are presented as an additional contribution to our knowledge of the habits of these two species. Frankly, my observations concerning the biology of the two Illinois species of *Psithyrus* are far from complete, but nevertheless they throw considerable light upon the habits of these inquilinous bees.

1. **Psithyrus variabilis** (Cresson)

I have found the adults of *P. variabilis* in eight nests of *B. americanorum* (Fabr.) (Fig. 3). In 1916, I published a short article in which I recorded the discovery for the first time in North America of adults of *variabilis* in the nests of *americanorum* in 1910 and 1915. Except for reference to this article by myself in 1921 and by Plath (1922a), no further mention of the habits of *variabilis* is given in the literature.

Almost all of the writers on the habits of the Bremidae have maintained that no workers of *Psithyrus* are produced. A discussion concerning the truth of this statement involves at the very outset a definition of *worker*. As this is discussed in an unpublished manuscript of mine it must suffice here to state that the workers are nothing but miniature females or queens which aid in colony production. If we grant that queens and workers are the result of a difference in feeding, small *Psithyrus* females "are comparable to *Bremus* workers" as Plath (1922a) has stated. In structure such small females of *Psithyrus* resemble the large females and hence can not

participate to any great extent in some of the duties performed by the small females or workers of *Bremus*. As we know so little about the behavior of small females of *Psithyrus* in the nests of bumblebees, it is well to speak of them as small females instead of queens.

Host

The fact that I have never found adults of *P. variabilis* in the nests of any species of bumblebee but *B. americanorum*, indicates that *americanorum* is the usual host of *variabilis*, in Champaign County, Illinois. Whether *americanorum* is the only host of *variabilis* or not, is another question, for it might be claimed that a sufficient number of nests have not been examined to prove that *americanorum* is the sole host of *variabilis*. The distribution of the two bees involved is such that it points to a similar relationship in other parts of the country. If *P. variabilis* infests the nests of other species of bumblebees it would be logical to expect to find the adults in nests of species closely allied to *B. americanorum*. The closest ally of *americanorum* in the states west of the Mississippi River is *B. fervidus* (Fabr.). More nests of this species of bumblebee have been examined than of any other North American form and none of these nests have revealed the presence of *Psithyrus*. Observations of Plath (1922b) also indicate that nests of *B. fervidus* are not susceptible to a *Psithyrus* infestation because of their peculiar "daubing" habits.

Hibernation

The large females of *P. variabilis* hibernate during the cold season. Tho no queens of this genus have hibernated under controlled conditions, the truth of this statement is based on conclusive data. In the first place, I have found a large female of *variabilis* hibernating in the decayed inner portion of an old stump on November 6. The queen was in the best of condition, altho in a lethargic state as a result of the cold weather. Without such proof as this, the fact that the adults are reared during the summer months in the nests of bumblebees, and that only females are found in the early part of the season, is sufficient evidence that at least some of the females hibernate as do their related *Bremus* hosts. Whether small females of *Psithyrus* hibernate, it is still an open ques-

tion. In this vicinity the females of *variabilis* which I have found in spring were all large.

Seasonal appearance of the adults

With the advent of warm weather the over-wintering females leave their hibernating quarters and frequent flowers. I have never found the females in spring to be very plentiful in the vicinity of Urbana, Illinois. In fact, I have never taken over five of them in one day. This is due partly at least to the fact that they are rarer than their host, and perhaps to some extent also because of their more reticent habits.

Over-wintering females of *P. variabilis* commonly appear about Urbana, Illinois, in the latter part of May. Many of the females are still found flying around in the early part of July. The lateness of their flight is no doubt due to their mode of living, which necessitates the finding of a bumblebee nest. The colonies of the host of *variabilis* are often well advanced by the first of July. The earliest date on which I have taken a female of *variabilis* in this vicinity is April 22, 1919, in the University Woods.

Apparently to the late establishment of the females in the nests of their host is due the fact that I have rarely found the males before August. The males are most commonly found on flowers in the latter part of August and September, and not infrequently in the early part of October. The earliest date on which I have taken a male is July 7, 1920, at Poy Sippe, Wisconsin. The statement that the progeny of the over-wintering females are usually produced late in the season is strengthened by the fact that in all cases where I have found *variabilis* in nests of *americanorum*, the nests were opened in September and October. Some of the females are seen flying around in the fall, and I have sometimes taken them on flowers along with the more numerous males. It always seemed to me, however, that they remained in the nest as long as possible and immediately after leaving the nest and possibly visiting a few flowers sought hibernating quarters.

Anthophilous habits

The fact that the females of *Psithyrus* do not forage for pollen and nectar for their young, accounts in a large measure for the rareness of records relating to the flowers visited by

these bees. Another factor is the almost total lack of flying *Psithyrus variabilis* adults in the field the last of July and first of August. The records are sufficient, however, to prove that *P. variabilis* is polytropic and not different in this respect from *Bremus*. The records are as follows: Cockerell (1899); *Prunus americana and Senico*. Swenk (1907): *Rubus*, *Verbena*, *Liatris*, *Bidens*, *Solidago*, and *Carduus*.

In addition to the above I have found the females on *Allium*, *Zinnia*, etc., and the males on *Eupatorium*, *Monarda fistulosa*, *Zinnia*, *Trifolium*, *Helianthus*, *Cornus*, *Aster*, etc.

Duration of the developmental stadia

No opportunity was afforded for a study of the duration of developmental stages in *Psithyrus*. The fact that the males and queens of *variabilis* are found in nests of *americanorum* in September, and the males in the field occasionally even in July, is sufficient evidence to indicate that the duration of the developmental stadia of *Psithyrus* does not greatly differ from that of *Bremus*.

Effect on colonies of Bremus

There has been considerable divergence of opinion as to what effect the presence of *Psithyrus* has on a colony of bumblebees. Only a statement as to the conditions which I found to exist in *americanorum* colonies infested with *variabilis* is given here, as a general discussion in regard to this topic is given in an unpublished manuscript article. In judging the effect the presence of any species of *Psithyrus* may have on a *Bremus* colony, several things must be taken into consideration, such as the probable time the colony of *Bremus* was established, time of invasion of colony by *Psithyrus*, infestation by other parasites and inquilines, situation of colony, date of nest examination, etc. In several colonies infested with *variabilis* examined in early fall, the old mother queen *americanorum* was still alive in the nest. This is quite conclusive evidence that the old *americanorum* queen is not always, if ever, killed by the invading female of *variabilis*.

Colonies of bumblebees may attain considerable size even tho infested with *Psithyrus*. One colony which I examined on September 17, contained three hundred and fifty cocoons

from which bees had emerged, beside fifty-eight cocoons containing larvae and pupae. In this nest were fifty-one adults of *americanorum*, one of which was a young queen. When this nest was first opened it contained but one female and three males of *P. variabilis* and I feel quite certain that few if any other *Psithyrus* adults had emerged. During the last of September and early part of October, however, thirty-six females and thirty males of *variabilis* emerged. It seems to me that the size of the comb and the number of worker bumblebees still alive in this nest, both indicate that this colony was not greatly handicapped by the situation of the nest, parasitism by insects other than *Psithyrus*, or thru late establishment of the nest in the fore part of the season. The emergence of the females and males of *variabilis* in numbers after September 17 is good evidence that few emerged prior to that time. In spite of the seventy adults of *Psithyrus* which emerged in this nest, this colony of *Bremus* attained considerable size and produced at least one young queen. Tho my notes do not state whether the empty cocoons were of workers, males, or queens, I believe that many queens and males of *americanorum* were produced in this colony and that they had left the nest before it was opened. Aside from the fact that one young queen was still in the nest, it should be understood that the majority of queens and males of *americanorum* in this vicinity emerge during the last of August and early part of September. Apparently, then, in this instance under many favorable conditions a fairly large colony of *americanorum* was produced in spite of a *varabilis* infestation. That the presence of *variabilis* hindered the greatest possible development of this colony of *americanorum*, it is also certain, for some seventy of these false bumblebees were reared. The food and time required of the bumblebees for the rearing of these *Psithyrus*, certainly represents the production of at least just that many fewer true bumblebees.

The study of two other colonies of *B. americanorum* containing *P. variabilis* proves that such colonies frequently produce males and queens of *Bremus*. It has been stated that in the case of an infestation of a *Bremus* nest by some species of *Psithyrus* no *Bremus* queens are henceforth produced. In one nest opened on September 14, I found nine males, thirty-eight workers, the old queen, and twenty young queens of

Bremus, besides one female *Psithyrus*. Evidently, males and queens of Bremus were produced in spite of the queen *Psithyrus*. Of course, it may be argued that in this instance the queen *Psithyrus* was but a recent lodger in the nest. Additional proof is furnished, however, by another nest which was examined on September 24. In this latter nest I found four males of *americanorum* besides eight females and two males of *variabilis* in additions to the bumblebee workers. Unless these males of *Bremus* were produced in the nest, their presence can not be satisfactorily explained.

It is pure speculation upon my part whether colonies of *B. americanorum* are ever completely exterminated thru the rearing of *P. variabilis*. Several factors again enter into such a speculation as this. In the first place, do the females of *variabilis* destroy all the eggs or young larvae of *americanorum* in the nests they invade or in which they reside? Unfortunately, I have never had an opportunity to prove or disprove that *variabilis* females destroy the eggs or small larvae of *americanorum*. Granting that eggs may be destroyed by the *variabilis* females, the presence of young queens and males of *Bremus* in infested nests in September is strongly indicative that some of them at least escape destruction. This seems still more evident when we consider that the females of *Psithyrus* in this vicinity are rarely encountered in the field in late July, an indication that most of them have found and invaded nests of *americanorum* or else perished. If females of *variabilis* invading nests of *americanorum* in summer destroyed all the small larvae and eggs, few bumblebees would be available for rearing the brood of *variabilis*. It is by no means improbable, however, that colonies of *americanorum* entered very early in the season by females of *variabilis* might come to an end, particularly if the habit of destroying the eggs of *americanorum* is very pronounced. Another source of danger to the colonies of *americanorum* lies in the possible killing of the queen in case of a struggle.

Number of adults produced in a colony

The largest number of *P. variabilis* that I have ever found in or reared from a colony of *B. americanorum* is seventy. This colony of *americanorum* was of fairly large size, which leads me to believe that it requires a fairly large colony of

americanorum to produce a large number of *variabilis*. In another colony of *americanorum* twenty-nine adults of *variabilis* were reared. In only one instance was the combined number of males and females of *variabilis* found in a nest larger than the number of true bumblebees. In this case, however, evidence was present that a great many more workers, males and queens of *americanorum* had been produced, but had left the nest or died before the nest was opened.

Caste ratio

The data in my possession are not sufficient to prove that more individuals of one sex are produced than of the other. The number of males found in two nests was about equal to the number of females. The habits of the males are such, however, that they are more apt to leave the nests than the females and accordingly many may be missing from a nest when a count is made.

Variation

The pubescence of the males of *P. variabilis* is more likely to exhibit a color variation than is that of the females. This may be partially due to the reduction in the amount of pubescence on the body of the females, but the males of the *Bremidae* in general are more subject to color variation than the females. The dorsum of the abdomen is the area most subject to color variation in the males of this species. I have seen males with all the dorsal abdominal segments nearly black and others with these segments entirely yellow. The average specimen, however, has the dorsum of the abdomen mostly black, except for a large amount of yellow on the fourth and fifth segments. Specimens of males with different amounts of black and yellow pubescence are found in the same nest. The females show a great deal less yellow than the males, but at times specimens are found with some touches of yellow on the sides of the fourth and fifth dorsal abdominal segments. This condition in the females from Illinois has led me to the conclusion that *Psithyrus bicolor* Cockerell is merely a variety of *Psithyrus variabilis* (1923).

Considerable difference in size exists among males in the same nest and its occurrence is explained in the same manner as the variation in the size of *Bremus* males. The females

that I have found in bumblebee nests in fall were properly referred to as large females. I have never seen small females of *variabilis* in the nests or collected them in the field, but this is not proof that they do not occur. In fact, when one considers the conditions that determine the production of the small workers of *Bremus*, it is probable that they do occur.

Behavior of the adults in the nest

In nests of *B. americanorum* found in the field containing adults of *P. variabilis*, I have never seen the queen or worker bumblebees show any animosity towards the adults of *variabilis*. This, however, does not signify that their invasion of the nest at the outset is not recognized and their presence resented. I have performed several experiments in an effort to determine what happens when a female of *variabilis* is first placed in a nest of bumblebees.

In one experiment a female of *variabilis* was placed in a small colony of *B. bimaculatus*. The workers immediately recognized her presence as shown by movements of their antennae, position taken on the comb, and the abrupt termination of the work in which they were engaged. A few minutes later, when the female *variabilis* crawled into the inner part of the nest, she was immediately attacked by the workers. In order to protect herself better from the attacks of the *bimaculatus* workers, the *variabilis* female drew all her legs up close against her body and remained almost motionless. Not desiring any casualties the *Psithyrus* was quickly removed.

In another experiment a female of *variabilis* was introduced into an artificial nest in which a queen of *B. americanorum* had been placed a few days before. As soon as the female of *varabilis* entered the nest proper she was viciously attacked with mandibles and stung by the *americanorum* queen. The *variabilis* female was then removed. In two other experiments, in which females of *variabilis* were introduced into small nests of *bimaculatus*, the *variabilis* female was not attacked.

It was interesting to observe the manner in which the *variabilis* female acted in one of these experiments. Tho the presence of the *variabilis* female was immediately noticed by the queen and workers of *bimaculatus*, none of the bumblebees left the comb to attack her. The *Psithyrus*, apparently recognizing the danger of her position, was extremely deliberate in

all her actions. Every forward movement she made was monotonously slow. Several times, as she advanced a little further into the nest a worker approached as if to attack her. Under such conditions the *variabilis* female continued to remain very calm, but her legs were drawn more closely against her body. Gradually she gained admittance upon the comb without a struggle, and within two hours after being placed in the nest the female *variabilis* could apparently crawl about the comb with impunity. This experiment was performed during the last of May.

Two days after this *Psithyrus* was placed in the nest something happened in the nest which caused me to suspect that the queen of *variabilis* was destroying some of the *Bremus* eggs. In two egg cells which contained three or four eggs on May 22, an examination made on May 23 revealed the presence of but a single egg. These cells were situated on that part of the comb where the female *variabilis* usually stayed, a fact which suggested that the *Psithyrus* had destroyed the other eggs and laid her own in the cells. The *variabilis* soon lost all interest in the nest and I removed her on May 30. During the entire time that this *Psithyrus* was in the nest (nine days), no fighting took place between the two species of bees. On May 30, however, I placed a *bimaculatus* queen and the female of *variabilis* in the same small bottle and in scrambling around trying to get out, a fight was started. In the struggle which ensued the *Bremus* queen was stung, and within five minutes was completely motionless.

Judging from my experiments and the state of affairs found in infested nests I believe that *Psithyrus variabilis* rarely kills the queen of *americanorum*. In fact, I am inclined to believe that she gets on just as peaceably as possible with her host. In case of a fight, she is well equipped by nature to maintain her own and the queen, some workers, or both, may be killed. No doubt at times a combat results differently and the bumblebees are the victor.

One of the secrets of the successful invasion of the nests of the aggressive *B. americanorum* seems to be the calm and deliberate attitude of the female of *P. variabilis* under all conditions. I can scarcely imagine a female of *variabilis* successfully establishing herself in a normal developing colony of *americanorum* if her movements were of a nervous type. The

workers of *americanorum* are exceedingly irritable and in normal colonies immediately attack and kill other bumblebees which are placed in their nests. A struggle once started would probably result in the participation of many of the workers then in the nest and the female *variabilis* would be in great danger of being killed. The slow deliberate movements of the female of *variabilis* in the nest of bumblebees is not altogether a temporary adjustment to conditions, for the same style of movement is characteristic of this bee on all occasions.

Frequent experiments have shown that both the males and females of *variabilis* can cut their own way out of the cocoons.

Mating

I have never seen males and females in copula, tho I have seen the males attempt to copulate with females under confinement. In several instances I have seen males of *Psithyrus variabilis* attempt to mate with queens of *Bremus*. The fertilization of the queens of *variabilis* probably occurs for the most part in September.

2. **Psithyrus laboriosus** (Fabricius)

This species is only rarely found in the vicinity of Urbana, Illinois, and accordingly but little opportunity has been presented for its study. I have never found live adults of *P. laboriosus* in nests of bumblebees examined in August and September. On two occasions, however, I have found the large females of *laboriosus* dead or dying in the debris surrounding the nests of bumblebees opened in July, and in 1921 I published a short note recording these facts. At the time this article was published, nothing was definitely known of the habits of *laboriosus*. In addition to the information presented in that paper, I have made some further observatons on a large female of *laboriosus* which was introduced into a colony (Experiment 23, 1919) of *B. americanorum*.

Experiment 23, 1919

A domicile buried in the ground in the University Woods on April 18, 1919, was found to be inhabited by bumblebees on June 20, 1919. On this latter date, the queen was absent from the domicile and therefore the nest was not disturbed.

It contained at that time a wax-pollen honey pot and a small mass of pollen on which were several sealed egg cells. When the nest was opened and removed on July 22, it contained a queen and four workers of *B. americanorum*, besides four empty cocoons, fifteen cocoons containing pupae, two groups of larvae, and five egg cells. In the grassy material near the entrance of the nest were the remains of a queen of *B. americanorum* and a large female of *P. laboriosus*. It was quite evident that these two queens had met their death in struggling for the possession of or admittance into the nest.

This colony continued to grow and much to my regret no specimens of *Psithyrus* were reared from any of the eggs, larvae, or cocoons which were found in the nest when it was removed on July 22. During the first and second weeks in August this colony was subjected to considerable handling, which resulted in the death of the queen and many of the workers. By way of experiment, a large queen of *P. laboriosus* was introduced into the colony on August 16. This female was collected at the Shades, Indiana, on June 29, 1919, and kept alive since its capture in an aquarium jar.

Because this queen was introduced into the nest in the evening, I did not have an opportunity to observe just what happened in the nest when the workers discovered the presence of the *Psithyrus*. Two dead workers, showing no trace of internal parasitism, were found in the nest box the next morning, and I have no doubt that they were stung to death by the female *laboriosus* during a fight. At the time the dead workers were discovered the *laboriosus* female was on the comb and showed a very great interest in the nest. Several times I observed her brooding on cocoons in much the same manner as do the *Bremus* queens. The workers at this time displayed no animosity towards her and it appeared as if they had accepted her domination. Several times the following day, however, I saw her seize one of the *americanorum* workers and give it a mauling, but in no instance was the worker killed. On the morning of this same day I noticed that one of the egg cells containing the eggs of *Bremus* had disappeared. In the evening, the female *laboriosus* appeared more excited about the nest than ever, and besides brooding over the cells, was observed constructing an egg cell of wax and pollen on the top of a cocoon.

On the next day, August 18, I noticed that this cell was sealed, and upon examination found it to contain eight eggs, all laid contiguous to one another and in a more or less horizontal plane. This egg cell was a little more rounded on top than those usually constructed by *americanorum*. A worker chanced upon this egg cell soon after I had opened it and immediately closed it again. Tho I did not see the female *laboriosus* lay these eggs, I am sure that they were laid by her, for she made the cell, there was no queen of *americanorum* in the nest, and workers seldom laid as many eggs at one time as were laid in this cell. Two days later, I found another new egg cell in the nest similar to the one just described. This cell contained ten eggs laid in the same manner as before. When the female *laboriosus* discovered this cell to be open, she immediately closed the cell as neatly and quickly as a true bumblebee. The same performance was repeated when she discovered that the egg cell she had first made was partially open. Her actions in these two instances satisfied me that she was responsible for the eggs which were laid after her introduction into the nest.

Due to a necessary absence, I had no opportunity to examine this nest again until September 16. During this time several adult workers of *americanorum* emerged from their cocoons, and nineteen bumblebees were still alive in the nest on the sixteenth. According to the observations very kindly made for me by Dr. R. D. Glasgow during my absence, no *Psithyrus* adults were reared, altho some of the *Psithyrus* eggs hatched. The female *laboriosus* was found dead in the nest on August 26.

Information concerning this species

Except for my short note in 1921, and the more recent articles by Plath (1922a, 1922c) and Bequaert and Plath (1925), there is nothing in our literature relating to the biology of *P. laboriosus* besides records of flower visits and the dates of the capture of adults. The first article by Plath is very illuminating and is the most important paper thus far published on the habits of the North American *Psithyrus*. A colony of *B. vagans* that Plath had under observation was entered by a female *laboriosus* on August 10. The colony contained at that time the "old queen, about 50 workers, and several males."

Plath observed the female of *laboriosus* tear "open a mass of wax containing small *Bremus* larvae, so that three of the latter rolled to the bottom of the nest box, leaving the remaining four widely exposed." He also saw this female construct an egg cell in which several eggs were laid. One adult female of *laboriosus* was reared from this nest (the other larvae were parasitized by *Melittobia*) and hence Plath considered *B. vagans* as a host of *laboriosus*. In 1919, on the basis of the geographical distriction of the *Bremidae* in Illinois, I expressed the belief that *P. laboriosus* was "an inquiline in the nests of *B. fervidus* or *B. vagans*." Plath gives a very interesting description of the manner in which the female *laboriosus* mauled the workers of *vagans* without injuring them and came to be accepted as one of the colony. This author also found that females of *laboriosus* sometimes lodged in colonies of *B. fervidus* and *B. bimaculatus*. The *fervidus* colonies protected themselves by daubing the *Psithyrus* with a fluid but the *bimaculatus* colonies never "seriously objected to these intruders."

Shortly after this first paper by Plath on North American *Psithyrus*, the same author (1922c) recorded the presence of *laboriosus* in the nests of *Bremus impatiens* (Cress.).

The recent paper by Bequaert and Plath (1925) gives additional data regarding the occurrence of *laboriosus* in the nests of *B. vagans* and *B. impatiens*. In addition, in this article two forms of *laboriosus* are recognized (the typical *laboriosus* of Fabricius and variety *citrinus* of Smith), and the interesting biological fact brought out "that each of these two forms shows a decided preference for a particular species of bumblebee." *P. laboriosus* is stated to generally breed in the nests of *B. impatiens* and *P. laboriosus* var. *citrinus* in the nests of *B. vagans*. Whether additional captures of *laboriosus* in the nests of bumblebees will substantiate such a selective host preference by the two color varieties remains to be demonstrated. Both varieties of *laboriosus* occur in Illinois, as also do the two species of bumblebees listed as hosts.

Host.

Plath (1922a) thinks it well to restrict the term *host* to "those species of *Bremus* in whose nests a given species of *Psithyrus* is known to breed successfully." This restriction of the term *host* is certainly desirable and is employed in this

article. The finding of dead or paralyzed females of *Psithyrus* in a nest of bumblebees, however, is certainly evidence of an attempted invasion on the part of the female, and may serve to indicate the host.

Plath has shown that *P. laboriosus* can successfully breed in the nests of *B. vagans* and *B. impatiens*, and hence represent its known hosts. Whether it has more than these two hosts is a question for the future to solve. Experiment 23, 1919, certainly indicated that *P. laboriosus* can breed in the nests of *B. americanorum*. In this experiment the female of *laboriosus* established herself in the nest, made egg cells, and laid eggs. The poor condition of this colony at the time is the only factor which seems to have prevented the rearing of the adults. None of my experiments or observations have shown that bumblebees can distinguish between the immature stages of *Psithyrus* and those of their own. Accordingly, larvae from eggs once properly laid in cells, other conditions being favorable, will be reared by the bumblebees. The fact that a female of *P. laboriosus* was found dead in the nest debris surrounding the comb in the early part of the season, strengthen the supposition that *laboriosus* can breed in the nests of *americanorum*. Of course, if we accept the term *host* as applying only to those species of bumblebee with which *Psithyrus* are known to have been reared, *americanorum* can not at this time be definitely listed as a host of *laboriosus*. The fact that I have also found a partially paralyzed queen of *laboriosus* in a nest of *auricomus* suggests that this latter bee may at times be a host of *laboriosus*, or that *laboriosus* is an occasional lodger in its nest.

Hibernation

The production of females in late summer and fall, their reappearance in spring after the cold weather, and biology which in many respects is similar to that of *Bremus*, all prove that females of *P. laboriosus* hibernate during the cold weather as do the queens of *P. variabilis*.

Seasonal appearance of the adults

The females are so rarely encountered about Urbana in spring, that I must rely on the data obtained by a study of pinned specimens in various collections to show the time of appearance of the over-wintering females. Data in my posses-

sion show that such females are usually collected in the field in May, June and July. Specimens are still flying in August and at this late date enter bumblebees' nests as shown by Plath. The lateness and prolonged season of the flight of these females are certainly due to their inquilinous habits. Females found flying in September and October are most likely all young females. The males are usually not encountered until after the middle of August. They are quite common in many parts of their range in September and are often found in October. The late appearance of the males and females is easily explained as a result of the inquilinous habits of the species.

Anthopilous habits

There are only a few records of the flowers visited by this bee, a condition resulting from the absence of a true worker caste and the inquilinous habits of the females. The following records are sufficient to prove that this bee is polytropic.

Robertson (1891–1896): *Asclepias verticillata* and *Ptelea trifoliata.*
Lovell (1907): *Pontederia cordata, Inula helenium, Carduus odoratus, Epilobrium augustifolium* and *Solidago.*
Bequaert (1920): *Cephalanthus occidentalis.*

In addition I have taken the males on *Eupatorium purpureum, Eupatorium perfoliatum,* and *Helianthus grosseratus.*

Duration of the developmental stadia

Plath (1922a) reports that a female of *P. laboriosus* emerged on September 7 from a cocoon spun by a larva about August 25. This larva had hatched and developed from an egg laid on August 10. This conclusively shows that the developmental period is quite similar to that found in *Bremus.* The fact that the larvae spun cocoons about fifteen days after the eggs were laid suggests an egg period of about four days, a larval period of about thirteen days, and a pupal stage of about ten days. The entire time from egg to adult required about four weeks.

Effect on colonies of Bremus

Since no colonies of *Bremus* have been found in the field which were infested with numbers of *P. laboriosus,* few data are available as to the effect of this bee on the development of a colony of bumblebees. Plath (1922a) in his first paper,

says that none "of the *Bremus* eggs and larvae—which were present in the nest when the *Psithyrus* queen gained admittance to the colony" developed to maturity. This does not prove, however, that bumblebees are not reared in nests so infested. It was not determined in this instance whether the *Bremus* queen laid any eggs after August 10. All *Psithyrus* adults, however, which are reared in a colony certainly represent just that many true bumblebees which might have been produced. This, if nothing else, is sufficient to cause *laboriosus* to be classed as an enemy to its host. No doubt at times workers and even the queen *Bremus* may be killed. The time of admittance of a female of *laboriosus* into a nest of *Bremus* has a great deal to do with the amount of harm that a *Psithyrus* female and her progeny can do.

Number of adults produced in a colony

Bequaert and Plath list five females and eighteen males as the largest number of *laboriosus* found in a single nest of *B. vagans*, and eighteen females and thirteen males as the largest number found in a nest of *B. impatiens*. The strength of the *Bremus* population in these nests was not stated.

Caste ratio

The data given by Bequaert and Plath obtained from the examination of bumblebee nests indicate a preponderance in numbers of the male sex. The same condition is reflected by my examination record of adult specimens in various collections, but of course such an index as to caste ratio is by itself unreliable.

Variation

P. laboriosus, based upon a consideration of the color of the pubescence, exhibits two varieties, the typical *laboriosus* (Fabr.) and *laboriosus* var. *citrinus* (Smith.) This division of *laboriosus* into two groups was first clearly defined by Bequaert and Plath. As already noted these authors also presented evidence to indicate a selective host preference on the part of the two groups. Specimens presenting transitional color phases between the two are not uncommon in collections, and both forms have been found in the same nest. Considerable variation in size exists in both sexes.

Behavior of the adults in the nest

Plath has recorded the mauling given to workers of *B. vagans* by a female of *P. laboriosus*, and I have seen workers of *americanorum* subjected to the same treatment. According to the same author, *P. laboriosus* can construct her own egg cells and my studies confirm this statement. In fact, I found that the *Psithyrus* female was as efficient in this respect as a queen of *Bremus*, and continued to care for these cells for some time after the eggs were laid. Brooding on the cells is not confined to *Bremus* queens, either, for I have seen the female of *P. laboriosus* so engaged. Evidently after the *Psithyrus* once becomes established in the nest, she remains on peaceable terms with the rightful owners.

B. Parasites of the Adults, Larvae and Pupae

1. **Physocephala sagittaria** (Say)

Adult bumblebees are frequently killed by the internal parasitism of the larvae of *Physocephala sagittaria* (Det. J. R. Malloch) (Fig. 2). The eggs of the fly are probably inserted by the female in the abdomen between the segments of adult bees while they are gathering nectar and pollen from flowers or working within the nest. Tho the puparia of this fly are frequently found in the abdomens of dead bees in the nests in late summer and early fall, I have not seen a record of this species being reared. In 1918, I reported finding the puparium of *Zodion obliquefasciatum* Macq. in the abdomens of dead females of *B. auricomus*. The rearing of *Physocephala sagittaria* in 1919 from similar puparia indicates that the 1918 record should be referred to this latter species. Records of parasitism by conopids of bumblebees are rather common in the European literature, the articles of de Meijere (1903, 1912) being the best available on the biology of these parasitic flies. Hoffer (1882–83) mentions that the larvae of *Conops* and *Physocephala* attacked the larvae and pupae of bumblebees and then emerged from the adults. The first part of Hoffer's statement is probably erroneous.

For some reason, I have found it exceedingly difficult to rear adults from *Physocephala* puparia and first succeeded in

so doing in 1919. Several conopid puparia were found in the abdomens of dead workers of *B. auricomus* on February 6, 1919. These dead workers were found in an old natural nest of *B. auricomus* which I examined in winter. One of these puparia produced an adult of *Physocephala sagittaria* on May 8, 1919. The fact that conopid puparia are commonly found in the abdomens of dead bumblebees in nests in late summer and fall, and the facts as just related, clearly show that *Physocephala sagittaria* hibernates as a puparium at the place where the infested bumblebee dies. The actions of bumblebees infested with conopid larvae are quite similar to those I have described for *Anthophora abrupta* Say (1922.) Packard (1864–65) reports having reared a species of *Conops* from an adult bumblebee. The specimen was in very poor condition and not easily determined and accordingly I believe there is now evidence to indicate the *Physocephala* was the genus involved in the parasitism.

The larvae of *Physocephala sagittaria* (Fig. 2a) are odd in appearance and when their anterior segments are extended remind one in some respects of a rat-tailed maggot, except that the cephalic and caudal regions are reversed. The living larva is creamy-white in color. The head and thoracic segments are somewhat cylindrical and only about one-fifth the diameter of the abdominal segments. The length of the full grown larva with the anterior segments extended is approximately 14 mm. The concave stigmal plates are very conspicuous and, depending upon the age of the larva, vary from a reddish-brown to a dark brown in color. A feature of the stigmal plates are the numerous wart-like protuberances and the presence on each plate of a somewhat centrally placed round pale spot or button. The stigmal plates are separated by much less than their smallest diameter.

The puparium (Fig. 2b) is about 8 mm. in length with a diameter of about 5 mm. It is reddish-brown or dark brown in color and is formed within the abdomen of the host bee. The stigmal plates still retain their peculiar appearance. The surface of the puparium which presses against the dorsal wall of the abdomen of the host is somewhat more arched than the lower or opposite surface.

Many conopid pupae from bumblebees produce a secondary parasite, *Mestocharis williamsoni* Girault (1911) (det. A. B.

Gahan). I have reared the same secondary parasite from the conopid parasites of *Anthophora abrupta*, tho I have never succeeded in rearing the conopid adult.

2. **Spherularia bombi** Dufour

Adult American bumblebees also die from the attacks of *Spherularia bombi* as recorded by Stiles (1895). I have frequently had queens in my observation boxes die in late spring and upon examination found the abdomen to be swarming with the immature forms of this curious parasite. I have found them in the adults of *B. vagans* and *B. americanorum*. Stiles records them from these two species and in addition *B. fervidus*. My species were provisionally identified for me from immature forms by Dr. N. A. Cobb. This parasite was originally described from Europe and according to Leuckart (1887) the fertilized female enters the body of a queen bumblebee and hibernates within its host. With the resumption of activity again on the part of the queen in spring the nematode undergoes a curious development and produces an enormous number of young. According to Stiles this parasite was first discovered by Leon Dufour in 1838, but I find that Reaumur in 1742 recorded finding millions of small worms in the abdomen of a female bumblebee and his description of them is such that it seems certain to me he was dealing with what we now call *Spherularia bombi*.

3. **Brachycoma sarcophagina** (Townsend)

The larvae and fresh pupae of *Bremus* (probably also *Psithyrus*) are killed by the attacks of a dipterous parasite—*Brachycoma sarcophagina* (det. J. R. Malloch) (Fig. 1). This parasite may create havoc with a bumblebee colony and with colonies under controlled conditions I have often been forced to use measures for its control. The adults invade the nests of bumblebees and by their agility evade the watchful bees. I have captured the adults in nests of various species and upon one occasion the captured adult immediately started to lay what I thought were eggs. An examination showed that the adult female had produced living young and accordingly this species of parasite must be considered as being at times at least viviparous. The habits of the adults and the fact that

bumblebee larvae are subject to attack indicate that the female adults of *Brachycoma* (Fig. 1a) deposit their young, or perhaps also eggs, on or in larval cells. Bumblebee larvae parasitized by this dipteron usually develop sufficiently to spin a thin silken cocoon when about to pupate. Soon after this the *Brachycoma* larvae (Fig. 1d) leave their host larva or pupa, which promptly collapses. The cocoons containing such parasitized forms have a spongy watery appearance. Worker bumblebees recognize that something is wrong within these thin, soft cocoons and open and renovate them. Several times I opened these cocoons before the workers and in such cases the *Brachycoma* larvae hurriedly crawled out of the cocoons thru the openings I had made and down onto the comb and into the nesting material. Under natural conditions no doubt the larvae of *Brachycoma* take advantage of the opening made in these cocoons by the bumblebees if it is made early enough, but it is probably an easy matter for them to escape thru the thin cocoons without outside aid. The larvae of *Brachycoma* after leaving the cocoons are undoubtedly sometimes encountered by the bumblebees and are then carried from the comb. Usually, however, the *Brachycoma* larvae work their way so quickly into the nest material and bottom of the comb that they escape the workers. In either case they are enabled to pupate successfully.

I have reared numerous adults of *Brachycoma sarcophagina* from puparia (Fig. 1b) and have found this species in the nests of *B. auricomus*, *B. fervidus*, *B. americanorum*, *B. bimaculatus* and *B. vagans*. Several or more generations may occur during the summer months, and experiments in rearing them show that the puparium stage may be as short as seven days. The larvae must likewise have a short period of development since they parasitize bee larvae which require about two weeks for development. I have reared adults from larvae and puparia found in my colonies the last of June and May (host *B. bimaculatus*) and the last of July and August (host *B, americanorum*.) This dipteron must hibernate either as an adult or as a puparium. Davidson (1894) in California reared a species of diptera from larvae found feeding on the larvae of *B. fervidus*, which was named *Brachycoma davidsoni* by Coquillet. According to Davidson, the *Brachycoma* larvae escape without outside interference from the cocoons and their eggs are laid

on the bumblebee larvae. *Brachycoma sarcophagina* has likewise been recorded by Plath (1922c) as a parasite of the larvae of *Bremus bimaculatus*. In England, *Brachycoma devia* is often a serious enemy of bumblebees according to Sladen (1912a).

I have captured the adults of *Phytoadmontia setigena Coquillet* (det. J. M. Aldrich) in the nests but did not obtain any accurate information as to their habits, tho it is likely that they are parasitic species.

4. **Mellitobia** sp.

Another internal parasite of the larvae and pupae of *Bremus* (also *Psithyrus*, see Plath, 1922a) is a species of *Mellitobia* (det. A. B. Gahan). Few data were obtained relating to the biology of these parasitic hymenopterous insects. Evidently the eggs are laid by the adult on or within the larval cells, and after the cocoon is spun the host succumbs.

5. **Parasitus** sp.

Adult bumblebees are frequently infested with mites of the family Parasitidae, genus *Parasitus* (det. H. E. Ewing). I have found these mites on many species of bumblebees, of all castes and from almost all regions. Sometimes they are so numerous that their presence is a serious menace to their host, but a few of them seem to cause no real harm. They are often found on queens in spring and this indicates that they probably hibernate on the bodies of queens and then, after the establishment of nests, breed within the nests. Sometimes the comb becomes badly overrun with them. Banks (1919) records *Parasitus bomborum* Oudemans from bumblebees collected by the Canadian Arctic Expedition of 1913–18. In answer to my inquiry Dr. Banks replied that it was very doubtful if all the specimens of this genus on *Bremus* were the same species and that there is possibly a group of allied species or even other species are involved. *Parasitus bomborum* was originally described by Oudemans (1902) from specimens taken on grass and on *B. terrestris* of Europe. In the arctic regions it is reported from willow catkins (Banks, 1919).

C. Insect Enemies of the Bumblebee Comb

1. **Vitula edmandsii** (Packard)

The most serious damage to the comb is caused by the attacks of *Vitula edmandsii* (det. J. McDunnough). This species of moth was originally described by Packard (1864) from specimens reared from larvae found in the nests of bumblebees. It has since been reported from bumblebee nests by Franklin (1913), and the writer (1918). According to my experience the larvae of this moth thrive equally well in the nests of any species of bumblebee, but particularly in weak colonies and after the colony becomes disorganized. Evidently the larvae of this moth hibernate within the silken tubes which they construct, for I have found the larvae of this moth in the nest debris of a nest of *B. americanorum* on October 3, 1920, and in an old nest of *B. auricomus* on February 6, 1919. In the laboratory some of the larvae found in the nest on October 3, pupated in January and moths emerged the same month, while others did not produce moths until the end of March. The larvae pupate within the tunnels in which they live. My observations indicate that several generations may be produced during the summer and I have reared adults from nests in June, July, August, September and October. Under natural conditions I have found moths and pupae in the nests as late as October.

The larvae of this moth according to my experience feed upon wax, pollen and the comb of bumblebees, but not on the live larvae or pupae. The larvae spin a great deal of silk which, together with the nesting materials to which it is attached, forms protecting tunnels or tubes. These tubes at times become very numerous and a regular labyrinth of them may surround the comb. The larvae remain within these tubes and by means of them are enabled to attack the bumblebee comb and elude the watchful bees. I may say, however, that bumblebees usually rear their brood in spite of them and that portion of the comb in most frequent use (the part with eggs, larvae and cocoons containing pupae) is seldom greatly damaged. The neglected part of the comb, and also the entire comb after its climax development and the males and queens are produced,

is speedily overrun with the larval chambers and soon is reduced to a dilapidated condition.

In Europe the larvae of a moth, *Aphomia sociella* Linnaeus, have been recorded by numerous investigators as occurring in the nests of bumblebees. According to Hoffer (1882–83) this is the worst enemy of the bumblebees and he says that the larvae will quickly ruin the comb, and besides eating the materials of which it is made eat the larvae and pupae. Sladen (1912) says practically the same thing. In the case of *V. edmandsii*, I have found no evidence that they will directly attack growing larvae or cocoons containing pupae while such immature forms are being properly cared for by the bees. The moment the colony becomes weakened, however, the greater the inroads made by the larvae of *V. edmansii*, and neglected larvae might be then destroyed.

The larvae of *V. edmansii* in turn are victims of at least two parasites, namely, *Habrobacon juglandis* Ashmead (det. R. A. Cushman) and *Apanteles nephopterícis* (Packard) (det. C. F. W. Muesebeck). I have reared the former from the larvae of *V. edmansii* which were found in a surface nest of *B. fervidus* on August 11 at Riverton, New Jersey, and from larvae taken in the nest of *B. impatiens* at Champaign, Illinois, in September. *H. juglandis* has been recorded from a fairly large number of hosts. The discovery of *Apanteles* (*Microgaster*) *nephopterícis* is very interesting since this species has not been previously rediscovered since it was originally described by Packard as parasitizing *Vitula* (=*Nephopteryx*) larvae from the nests of bumblebees. In a recent paper by Muesebeck (1921) the *Microgaster nephopterícis* of Packard had to be considered as questionably belonging to *Apanteles*. The rearing of specimens of a parasite agreeing quite well with Packard's original description of *Microgaster nephopterícis* and from the same host suggested that here was possibly an opportunity to clear up the status of Packard's doubtful species. Mr. Musebeck very kindly studied this material and wrote me that the specimens belonged to *Apanteles* and were what was described by Packard in 1864–65 as *Microgaster nephopterícis*. Since Packard's description is very poor the following redescription of this species by Mr. Muesebeck is included.

Apanteles nephoptericis (Packard)

(Redescribed by C. F. W. Muesebeck)

"Many specimens, both sexes, reared by T. H. Frison from (*Nephopteryx*) *Vitula edmandsii*, Champaign, Illinois.

"Female.—Length 3 mm. Antennae distinctly shorter than the body; face, vertex and temples minutely punctate, shining; mesoscutum opaque, entirely very closely punctuate; scutellum shining, but distinctly punctuate; propodeum with a moderately broad median areola, which is not prominently margined anteriorly but is closed; propodeum mostly rugulose, smooth and shining in the posterior lateral angles; mesopleurae closely punctuate on anterior half, polished posteriorly; fore wing with large broad stigma; metacarpus no longer than stigma; radius slightly longer than the intercubitus, and somewhat directed outward, with the suggestion of a heel at the point of union with intercubitus; posterior coxae weakly punctate, not polished; first dorsal abdominal plate parallel-sided, rugulose, with a smooth and shining longitudinal fovea on posterior half; second plate transverse, with the posterior margin arcuate, mostly smooth and shining, being weakly striate only along posterior margin; remainder of abdomen smooth and polished; ovipositor sheaths very nearly as long as the abdomen. Black; antennae wholly black; tegulae yellow; wing-bases brown; costa yellow; stigma pale brown with a large pale spot at base; dorsum of abdomen entirely black, including the membranous margins along basal plates.

"*Male.*—Antennae as long as the body; otherwise agrees well with female."

2. **Plodia interpunctella** Hübner

The larvae of *Plodia interpunctella* breed very rapidly in the comb of bumblebees and with colonies under controlled conditions their attacks must be guarded against. Davidson (1894) has reported finding *Ephestia kuehniella* Zeller in the nests of *B. fervidus* in California.

D. Scatophagous Associates and Occasional Lodgers

A large number of other insects act as scavengers within the bumblebee nest. Among the most interesting of these is the beetle *Antherophagus ochraceus* Melsheimer (det. C. A. Frost). Many European investigators record the capture of *Antherophagus* (*pallens*, *silaceus* and *nigricornis*) in the nests of bumblebees and some of them have reported finding the adult beetles attached to the proboscis of bumblebees. A similar habit of attaching itself to the proboscis of a bumblebee has been recorded by Wheeler (1919) for the American *Antherophagus ochraceus*. This beetle and its larvae were first found by Packard (1864–65) in the nests of bumblebees and later by myself (1921b.) Since then Plath (1922c) has found the adults and larvae in the nests of bumblebees.

The adults gain admittance to the nests of bumblebees often if not always by attaching themselves to the proboscis of bumblebees while the latter are feeding at flowers. They are then carried back to the nests and upon releasing their hold fall onto the comb and eventually reproduce their kind in the debris of the nest, particularly in that at the bottom of the comb. For a more detailed account of the "phoresy" and the habits of *Antherophagus ochraceus* the reader is referred to the papers of Wheeler (1919), Donisthorpe (1920) and my own (1921b). The larvae certainly are not predaceous upon the young of bumblebees, and according to my observations this beetle is incapable of causing an enormous devastation of the comb as Wagner (1907) has stated it is the case with the European *Antherophagus*. After the comb has been abandoned by the bumblebees, the larvae of this beetle no doubt aid its disintegration. I have taken specimens of the larvae and adults of this species of beetle in the nests of many species of bumblebees.

I have also taken the adults of *Crytophagus croceus* Zimmerman in the nests of *B. americanorum* at White Heath, Illinois. The presence of the adults of European species of *Cryptophagus* in the nests of bumblebees has been recorded but I am not aware of any American records to this effect. The close relation of this beetle to *Antherophagus ochraceus* is suggestive that perhaps this beetle may gain access to the nests of bumblebees by phoresy.

Upon one occasion I found numerous larvae of *Tenebrio tenebrioides* Beauvois and *Tenebrio obscurus* Say (the adults were reared) which were acting as scavengers in the debris beneath the comb. Many other beetles undoubtedly at times lodge in bumblebee nests and I have found specimens of *Onthophagus hecate* Panzer, *Glischrochilus fasciata* (Oliver), and several small undetermined forms in nests. A very large number of beetles have been recorded from bumblebee nests in Europe, and the lists of Tuck (1896, 1897) are particularly comprehensive.

The larvae of Diptera often act as scavengers in the nests of bumblebees. The spiny larvae of *Fannia scalaris* Fabricius (det. J. R. Malloch) are very numerous in their nests and I have frequently reared them. Packard (1864–65) mentions the occurrence in nests of bumblebees of the larvae of an unknown dipteron which he thought were allied to *Volucella*. His figures indicate these larvae were those of *Fannia*. I have also reared the adults (all very kindly determined for me by Mr. J. R. Malloch) of the following flies from larvae acting as scavengers in bumblebee nests: *Muscina stabulans* (Fallen), *Drosophila busckii* Coquillet, *Aphiochaeta aletiae* Comstock, and *Scatopse strata* Say. Tho I did not have an opportunity to rear the adults I once found twenty-one larvae of *Volucella* sp. (probably *bombylans fascialis* Williston) in a nest of *Bremus fervidus* opened on August 12, 1920, at Baraboo, Wisconsin. These larvae lived in the nest debris beneath the comb and are certainly to be considered as scavengers. I feel certain they never molest the brood as has been stated, because their position in the nest, movements, and condition of the colony at the time preclude such a presumption. The citation of this syrphid as an example of aggressive mimicry, as has been done in the past, is erroneous. Plath (1922c) also found the larvae of *Volucella* in a nest of the same species of bumblebee.

Ants occur in bumblebee nests and several times I have encountered in the debris under the comb the adults of *Solenopsis molesta* Say (det. W. M. Wheeler) and *Lasius niger* var. *neoniger* Emery (det. W. M. Wheeler). Plath (1922c) has found the pseudoscorpion *Chelanops sanborni* Hagen in underground nests and suggests they may get into the nests by phoresy.

E. Miscellaneous Enemies

There is to my knowledge no American records of a case of microsporidiosis of bumblebees. I have had bumblebees die frequently in my observation colonies without showing any traces of any of the larger endoparasites, and I do not doubt now but that a more critical examination of these bees at the time would have resulted, at least in some cases, in the finding of microsporidia. *Nosema bombi* Fantham and Porter is known to occur in the alimentary canal and Malphighian tubules of several European species of bumblebees. A related species, *Nosema apis* Zander, often causes a serious disease of honeybees and experimental infections have been produced in bumblebees. The possibility of honeybees also contracting a *Nosema* disease from bumblebees is not to be disregarded. Bumblebee larvae often die from what seems to be a bacterial disease. How closely this disease is related to the bacterial diseases of the honeybee remains to be demonstrated. The entire subject of the diseases of bumblebees is in need of a thoro investigation and it would not be surprising if the results obtained affected some of our honeybee legislation.

The predaceous enemies of bumblebees are numerous. Upon several occasions in Illinois I have seen specimens of the large asilids, *Dasylis* and *Promachus*, with bumblebees in their grasp. Records of such instances are likewise found *in litt.* Among the vertebrate series the shrews, moles, gophers and mice are responsible for some destruction of bumblebee colonies, particularly at an early stage of their development. In 1919, I lost two incipient colonies of bumblebees due to the work of mice. According to a report made to me by Mr. D. H. Thompson, skunks destroy bumblebee nests. Mr. O. E. Plath (1923) has published an interesting article concerning the "Bee-eating Proclivity of the Skunk." Tho I have not found any records in the literature, a few bumblebees are probably eaten by Illinois birds. It is well known that the kingbird (*Tyrannus tyrannus* Linnaeus), abundant in Illinois, kills honeybees and there is no good reason for not supposing that it occasionally kills small bumblebees.

Man both intentionally and unintentionally is a decided factor in the diminuation of the bumblebee population. Intentionally, colonies are destroyed thru ignorance and for the honey which is stored in the wax-pollen cells and empty cocoons.

Unintentionally, the nesting places preferred by the bumblebees are destroyed by cultivation, deforestation, etc., particularly in areas of great agricultural activity such as Central Illinois.

F. Tabulation

In this tabulation is given a complete list of the known enemies of the bumblebees of Illinois and of the various inhabitants of varied status thus far found within their nests. There is no doubt but that continued investigations will reveal a few additional major enemies, and greatly augment the list of scavengers and occasional lodgers.

Social parasites:
- *Psithyrus variabilis* (Cresson). Host: *Bremus americanorum* (Fabricius).
- *Psithyrus laboriosus* (Fabricius). Hosts: *Bremus vagans* (Smith) and *impatiens* (Cresson).

Entoparasites of the adults:
- *Physocephala sagittaria* (Say). Causes death. *Mestocharis williamsoni* Girault is a secondary parasite.
- *Spherularia bombi* Dufour. Causes death.

Ectoparasites of the adults:
- *Parasitus* species (possibly *P. bomborum* Oudemans). Seldom injures host. Also found upon the comb and reported from vegetation.

Entoparasites of larvæ and pupæ:
- *Brachycoma sarcophagina* (Townsend). Causes death.
- *Phytoadmontia setigena* Coquillett. (Parasitism not conclusively established).
- *Mellitobia* species. Causes death.

Destructive to the comb:
- *Vitula edmandsii* (Packard). May ruin the comb, particularly when neglected. Larvæ parasitized by *Habrobracon juglandis* Ashmead and *Apanteles nephopterícis* (Packard).
- *Plodia interpunctella* (Hubner). Bred from nests under observation in the laboratory.

Scatophagus associates:
- *Fannia scalaris* Fabricius.
- *Tenebrio obscurus* Say.
- *Tenebrio tenebrioides* Beauvois.
- *Antherophagus ochraceus* Melsheimer. Adults carried into nests by adult bees—example of phoresy.
- *Crytophagus croceus* Zimmerman.
- *Muscina stabulans* (Fallen).
- *Drosophila busckii* Coquillett.
- *Aphiochaeta aletiæ* Comstock.
- *Scatopose atrata* Say.
- *Solenopsis molesta* Say.
- *Lasius niger* var. *neoniger* Emery.
- *Glischrochilus fasciatus* (Oliver).

Occasional lodgers:
- *Onthophagus hecate* Panzer. (Scavenger?).
- *Gryllus assimilis* var. *pennsylvanicus* Burmeister.
- Numerous small Coleoptera.

Miscellaneous enemies:
- Diseases.
- Predaceous insects.
- Some vertebrates, including man.

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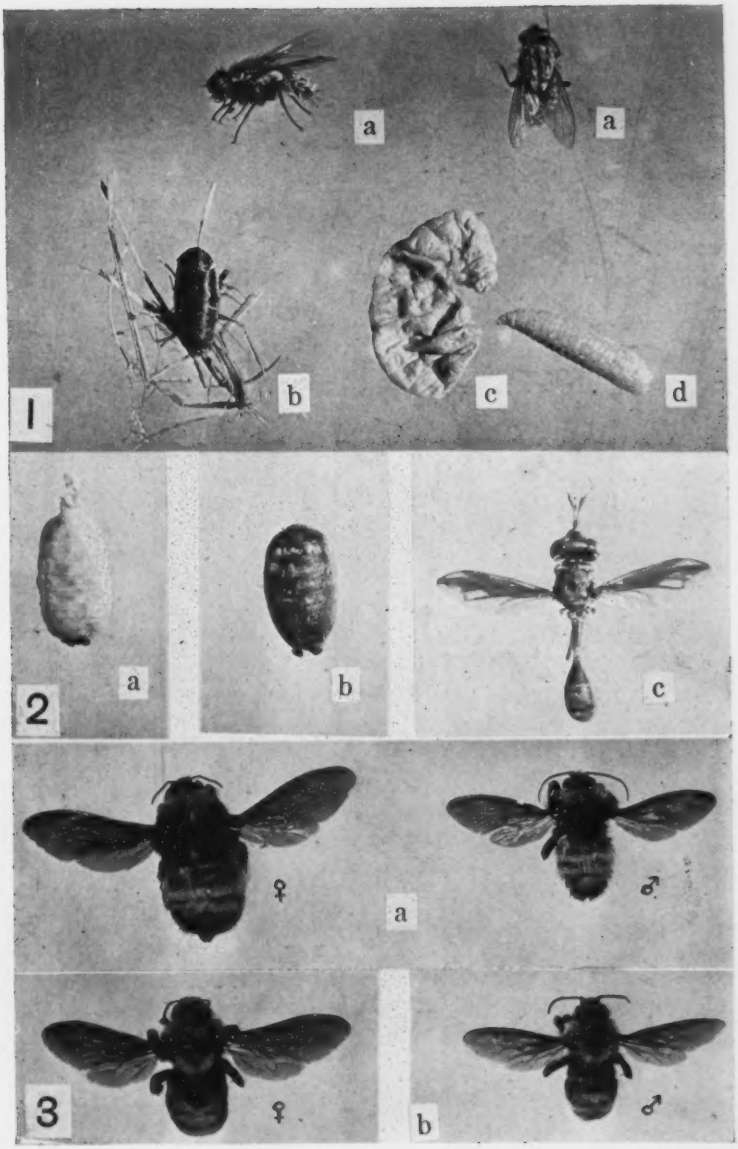
EXPLANATION OF PLATE XVII.

Fig. 1. A tachinid parasite of bumblebee larvæ, *Brachycoma sarcophagina* (Towns.). *a*, adult flies; *b*, puparium; *c*, bumblebee larva after emergence of tachinid larvæ; *d*, tachinid larva after leaving host.

Fig. 2. A conopid parasite of adult bumblebees, *Physocephala sagittaria* (Say). *a*, larva removed from abdomen of bumblebee; *b*, puparium formed in abdomen of bumblebee; *c*, adult fly.

Fig. 3. True bumblebee and its social parasite belonging to the same family of bees. *a*, true bumblebee, *Bremus americanorum* (Fab.); *b*, social parasite, *Psithyrus variabilis* (Cress.).



T. H. Frison.

CHARACTERS OF FAMILY AND SUPERFAMILY SIGNIFICANCE IN THE MALE GENITALIA OF MICROLEPIDOPTERA.

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The importance of the male genitalia in the classification of the Microlepidoptera has been discussed by Busck and Heinrich in 1921. Reference was then made to the supreme importance of these structures in the differentiation of species, and in the clearer and sharper definition and finer division of families and genera. It is the purpose of this paper to extend the use of the male genitalia to the characterizing of superfamilies and suborders and to the determining of the relationships of their constituent families. Following the suggestion of Busck, (1914), the Microlepidoptera is regarded as a natural group altho the families, as used by present day specialists, are not phylogenetically equivalent, many of the more specialized families having been derived from generalized ones of more than family rank. Because of this the Hepalidae and Micropterygidae, and their allies are regarded as superfamilies on a par with the Tineoidea and the discussion in this paper is limited to these three groups. The terminology used to describe the parts of the male genitalia is that already explained by the author, (1924) in a previous paper.

It would be folly to attempt a classification of the Microlepidoptera on the male genitalia alone. However a comparison of these structures in the families and larger units into which the group has been divided will assist materially in the ultimate establishing of a system of classification based on all the characters available in the group. A number of systems of classification have been proposed of which those of Comstock, Packard, Spuler, and Tillyard are summarized in the following table.

SCHEMES OF CLASSIFICATION FOR THE MICROLEPIDOPTERA

COMSTOCK (1893).

Suborder: JUGATÆ.
Macrojugatæ—Hepialidæ.
Microjugatæ—Micropterygidæ.

Suborder: FRENATÆ.
Microfrenatæ—Tineina.
Tortricina.
Pyralina.

PACKARD (1895).

Suborder: LACINIATA.
Micropterygidæ (in part).

Suborder: HAUSTELLATA.
Paleolepidoptera (pupæ liberæ).
Micropterygidæ (in part).
Neolepidoptera.
I. Tineoids (pupæ incompletæ).
(a) Generalized:
Prodoxidæ, Nepticulidæ, Tineidæ, Gracilariidæ.
(b) Specialized:
Remaining Tineina and the Hepialidæ.
II. Macrolepidoptera.
(pupæ obtectæ).

SPULER (1899).

TINEINA.
I. Aculeatae.
Incurvariidæ.
Incurvariinæ.
Adelinæ.
Phylloporiinæ.
Crinopteryginæ.
Eriocottinæ.
Heliozelidæ.
Nepticulidæ.
Nepticulinæ.
Oposteginæ.
Tischeriidæ.
II. Nonaculeatae: remaining Tineina.

TILLYARD (1919).

Suborder: HOMONEURA.
Hepialoidea—Hepialidæ.
Prototheoridæ.
Anomosetidæ.
Paleosetidæ.
Micropterygoidea—Micropterygidæ.
Eriocraniidæ.
Mnesarchaeridæ.

Suborder: HETERONEURA.
Tineoidea, Tortricoidea.
Pyraloidea and Macrolepidoptera.

The classification of Comstock, (1893), is of interest because he was among the first to separate the exceedingly primitive families Hepialidae and Micropterygidae from the remaining Lepidoptera on the basis of the wing coupling apparatus. The system of Packard, (1895), also recognized the generalized nature of Micropteryx in the establishing of his suborder Laciniata, and his division of the Tineina into a generalized and specialized group foreshadowed Spuler's more natural divisions, Aculeatea and Nonaculeatae in 1899. Tillyard, (1919), has suggested the names Homoneura and Heteroneura for Comstock's suborders Jugatae and Frenatae and has elevated the Hepialidae and Micropterygidae to superfamily rank including among them certain recently described families from the Australian and African fauna.

The male gentalia of the Homoneura, (Plate XVIII, Figs. 1–9) are characterized by the rudimentary condition of the aedoeagus and the articulation of the harpes to the juxta. In most Lepidoptera the aedoeagus occurs typically as a hollow chitinized sheath which forms the covering of the distal end of the ejaculatory duct or penis. In the Homoneura it may be entirely absent, as in the Mnesarchaeidae, (Plate XVIII, Fig. 9), represented by a chitinized plate or process as in the Hepialidae and Mnesarchaeridae, (Plate XVII, Figs. 3 and 8), or by a weakly or incompletely chitinized tube as in the Micropterygidae, (Plate XVIII, Figs. 6 and 7), or Eriocraniidae, (Plate XVIII, Fig. 5.) The harpes are entirely or in part articulated to the juxta and rarely show the close association with the vinculum which is characteristic of the harpes of the Heteroneura.

These two structures serve further to distinguish the two superfamilies of the suborder, i. e., the Hepialoidea and Micropterygoidea. In the Hepialoidea the aedoeagus is in the form of a chitinized plate which supports the membranous penis from the ventral side. This plate may be armed with one or more processes or spines. The aedoeagus of the Micropterygoidea is, as before mentioned, weakly chitinized or absent. When developed it is tubular, never plate like as in the Hepialoidea. In the Hepialoidea the juxta is broad and the harpes are widely separated at their points of articulation with it; in the Micropterygoidea it is narrow and the harpes are approximate or contiguous at their bases. The uncus and gnathos are rarely present and the former when it occurs is solidly fused to the tegumen, not articulated to it as is the uncus of most Heteroneura.

The families comprising the *Hepialoidea* and *Micropterygoidea* are listed below with a summary of the male genitalia characters which distinguish them.

Hepialoidea:

Hepialidæ—(Plate XVIII, Figs. 1 and 3). Eighth sternite heavily chitinized, forming a plate beneath the vinculum; vinculum usually large, V- or U-shaped; aedoeagus consisting of a small plate sometimes armed with a terminal process; juxta chitinized.

Prototheoridæ—(Plate XVIII, Fig. 2). Eighth sternite not plate-like; vinculum small, V-shaped; aedoeagus armed at the termen with large lobate and hooked processes; tegumen with two pairs of large process; juxta lightly chitinized.

Paleosetidæ—(Plate XVIII, Fig. 4). Eighth sternite not plate-like; vinculum small and U-shaped; aedoeagus a short process fused to the chitinized juxta; processes of the tegumen very elongate; harpes deeply divided into a sacculus and cucullus. A similar division of the harpes occurs in certain Hepialidæ, but this condition is exceptional and is not so pronounced as in Paleoses.

Anomosetidæ—Not examined.

Micropterygoidea:

*Mnesarchaeidæ—(Plate XVIII, Figs. 8 and 9). Eighth sternite normal; vinculum large and U-shaped; aedoeagus membranous; juxta membranous, sometimes armed with spines; tegumen armed with one or two pairs of processes.

Eriocraniidæ—(Plate XVIII, Fig. 5). Eighth sternite normal; vinculum large with the front margin often emarginate; aedoeagus with chitinized dorsal and ventral surfaces and membranous lateral ones; juxta a narrow chitinized bridge which connects the extremely small harpes; tegumen with one pair of lobate processes.

Micropterygidæ—(Plate XVIII, Figs. 6 and 7). Eighth sternite vestigial, having been crowded out by the enormous vinculum; aedoeagus a lightly chitinized tube foreshadowing the type found in the Heteroneura; juxta a small plate or rod which joins the bases of the harpes; tegumen simple or with a pair of lobate processes or a solidly-fused uncus.

The male genitalia of the Heteroneura differ from those of the Homoneura mainly in the chitinized tube-like aedoeagus and the articulate uncus and gnathos. The development of these structures is quite gradual, especially in the aculeate Tineoidea with the result that a membranous aedoeagus persists in the Opostegidae, the uncus, when present, is usually solidly fused to the tegumen resembling the type found in the Micropterygidae, and the gnathos is absent in the more generalized Aculeatae, i. e., Incurvariidae and Prodoxidae. In the Tineidae and higher Tineoidea these structures occur with remarkable regularity and, altho often highly modified or occasionally lost, may be regarded as the outstanding characteristics of the nonaculeate Tineoidea.

The Aculeatae differ from the nonaculeate Tineoidea in the retention of a large U or V-shaped vinculum, similar to that of the Micropterygoidea, and by the shortened tegumen which in very rare cases is terminated by an uncus. Figures 10–13, on Plate XVIII illustrate the principle modifications of the type of genitalia characteristic of the Aculeatae. Certain transitional types between these just described and those which characterize the Tineidae are found in such forms as Crinopteryx, Eriocottis and Talaeporia. In Talaeporia, (Plate XVIII, Fig. 15), the vinculum is smaller than in the Aculeatae,

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* Mr. Alfred Philpott, of the Cawthorn Institute of Scientific Research, has recently analyzed the male genitalia of the New Zealand Mnesarchaeidæ and has called my attention (in litteris) to several mistakes in my interpretation of these structures in this family. The "membranous plate" which I considered the juxta in my description in 1924 is the central plate of a "suspensorium" type of aedoeagus similar to that described for certain species of Hepialus and Sthenopis. The spines associated with this structure are the "cornuti" which are borne on the membranous penis. The structures described in *M. hamadelpha* Meyr., and *M. loxoscia* Meyr., as the "outer pair of finger-like processes of the tegumen" are the cuculli of the harpes which have assumed a dorso-lateral position and are obscured by the sacculi unless the harpe is removed. This type, of divided harpe, resembling closely that of the Paleosetidæ, occurs in all of the known species of the family except *M. fusca* Philp., (Plate XVIII, Fig. 9). Here the harpes are reduced and remind one of those in the Eriocraniidæ. Considering these changes in interpretation, the genitalia of the family are intermediate in structure between those of the Hepialoidea and Micropterygoidea and show closest similarity to the genitalia of Paleoses.

a short saccus is developed, the harpes are slightly divided while the tegumen remains large and simple very much as in the generalized Micropterygidae, i. e., Sabatinca, (Fig. 6.) Tinea xystidophora Meyr., (Plate XVIII, Fig. 14), represents one of the generalized Tineidae slightly in advance of the type just described in which the harpes and vinculum remain simple but the tegumen develops an uncus. From such generalized types have resulted on one hand the Tineinae, (Plate XVIII, Fig. 17), with an articulate uncus and gnathos and a vinculum with a highly developed saccus and, on the other hand, the Acrolophidae (Plate XVIII, Fig. 16) with highly specialized divided harpes, articulate uncus and gnathos and a simple vinculum.

The remaining families or higher *Tineoidea* as they may be called to distinguish them from the *Aculeatea* and *Tineidae*, are exceedingly diverse in genitalia structure. Some are modifications of the two types already described for the *Tineidae* and *Acrolophidae* while others depart quite widely and exhibit structural peculiarities of decided individuality. No attempt is made in this short paper to make a complete survey of them. A typical example of each of the more important North American families is figured along with a brief enumeration of the characters which serve to distinguish it.

Aculeate Tineoidea:

Prodoxidæ—Vinculum V-shaped; juxta membranous; aedoeagus slender and attenuate; uncus and gnathos absent.

Incurvariidæ—(Plate XVIII, Fig. 13). Vinculum U- or V-shaped; juxta membranous or lightly chitinized; aedoeagus short and heavy; harpes occasionally with short sacculus; gnathos absent; uncus occasionally present.

Adelidæ—(Plate XVII, Fig. 11). Vinculum and juxta as in Incurvariidæ; aedoeagus slender and attenuate; harpes often adorned with spinose or scobinate pads; uncus absent; gnathos occasionally present.

Heliozelidæ—(Plate XVII, Fig. 10). Vinculum U-shaped, exceedingly long; juxta membranous or lightly chitinized; anellus spined; aedoeagus long and slender, but not attenuate; harpes finger-like with one pair of spinose or scobinate pads, often petiolate; uncus absent; gnathos present.

Opostegidæ—Vinculum broadly U-shaped or emarginate; juxta membranous; aedoeagus membranous or lightly chitinized; harpes finger-like with one pair of spinose pads which are often large and complicated; uncus absent; gnathos often present.

Nepticulidæ—(Plate XVIII, Fig. 12). Vinculum U-shaped, rectangular, or emarginate; juxta membranous; aedoeagus very heavy and well chitinized; gnathos present; uncus often present.

Crinopterygidæ—Vinculum U-shaped; juxta well chitinized; anellus spined; aedoeagus short and heavy; harpes with a short sacculus; uncus absent; gnathos rudimentary.

Nonaculeate Tineoidea:

Talaeporiidæ—(Plate XVIII, Fig. 15). Vinculum U-shaped, with or without a short saccus; juxta chitinized; aedoeagus fairly heavy and well chitinized; harpes with or without sacculus, uncus absent; gnathos absent or rudimentary.

Tineidæ, (exclusive of the Amydriinæ and Scardiinæ)—(Plate XVIII, Fig. 17). Vinculum U- or V-shaped, usually with a long saccus, or occasionally quadrate; juxta chitinized or membranous; aedoeagus short and heavy, or long and slender, usually attenuate and occasionally elbowed, often enlarged at the base; harpes usually undivided; uncus and gnathos usually present.

Acrolophidæ, (including Amydriinæ and Scardiinæ)—(Plate XVIII, Fig. 16). Vinculum U-shaped, without saccus; juxta membranous or chitinized; aedoeagus large, dilated at base, usually with terminal spines or teeth; harpes divided into two or three distinct lobes or into a sacculus, clavus and cucullus.

Plutellidæ (and Yponomeutidæ)—(Plate XIX, Fig. 1). A pair of hair or scale tufts present on the intersegmental membrane of the 8th sternite; vinculum U- or V-shaped, usually with saccus; eighth sternite usually divided on the meson, forming a hood around the vinculum and harpes; transtilla well developed; uncus emarginate, bifed or with socii; gnathos present.

Gracilariidæ—(Plate XIX, Fig. 4). A pair of hair or scale tufts usually present on the intersegmental membrane of the 7th and 8th sternites; vinculum U- or V-shaped, often with a saccus; aedoeagus linear; tegumen weakly developed, often fused mesally with the anus; uncus and gnathos usually absent.

Lithocolletidæ—(Plate XIX, Fig. 11). Eighth sternite usually flap-like, forming a ventral hood; hair tufts rarely developed; harpes often unsymmetrical, divided or with irregular processes; aedoeagus attenuate or elbowed; tegumen as in the Gracilariidæ.

Lyonetiidæ—(sens. str.)—(Plate XIX, Fig. 6). Vinculum U- or V-shaped, often with a chitinized ridge on the median line; juxta membranous, usually indistinct from the anellus; aedoeagus attenuate, occasionally elbowed; harpes finger-like or occasionally divided into a finger-like and spoon-shaped portion; tegumen hood-like, without a gnathos; uncus absent or vestigial; socii usually present. The Old World Cemiostomidæ and Opogona do not conform to these characters and represent a distinct family or a more generalized branch which seems closely connected with the Crinopterygidæ and Tineidæ, but rather isolated from the more highly specialized Lyonetiidæ and Bucculatrix.

Heliodinidæ—(Plate XIX, Fig. 12). Scale tufts often present on the intersegmental membrane of the 8th sternite; Vinculum with an exceedingly long saccus; aedoeagus very long; harpes slender with petiolate bases; uncus and gnathos usually absent, socii present.

Elachistidæ—(Plate XIX, Fig. 2). Juxta heavily chitinized and with one or two lateral processes, transtilla exceedingly well developed; gnathos spined; uncus, when present, broad and flap-like. In some species of Cygnodia the uncus is absent and the gnathos replaces it as in the Coleophoridæ.

Coleophoridæ—(Plate XIX, Fig. 5). Vinculum V-shaped with the central portion weakly chitinized; harpes divided into a distinct sacculus and cucullus, uncus absent, but replaced by the large, heavily spined gnathos.

Blastobasidæ—(Plate XIX, Fig. 8). Vinculum narrow; aedoeagus short, weakly chitinized mesally; harpes divided into a distinct sacculus and cucullus; transtilla well developed; uncus and tegumen tending to become short and broad, especially in Pigritia.

Stenomidæ—(Plate XIX, Fig. 13). Vinculum U-shaped or rectangular with central portion weakly chitinized or narrow; juxta often with a pair of lateral processes; aedoeagus short and broad; harpes spoon-shaped, entire or divided and armed with spatulate, toothed or split hairs; uncus usually long and slender with the gnathos articulated close to its base by two slender arms.

Oecophoridæ—(Plate XIX, Fig. 3). Vinculum U- or V-shaped, occasionally with a short saccus; juxta usually well developed and adorned with two lateral appendages; aedoeagus large; harpes spoon-shaped, often divided into sacculus and cucullus; gnathos and uncus well developed.

Momphidæ—(Plate XIX, Fig. 14). Characters much as in the Oecophoridæ; harpes more distinctly divided; vinculum broader and occasionally with a saccus. Erineda, Mompha, Psacaphora, and possibly Epermenia and Schreckensteinia conform with these characters.

Cosmopterygidæ—(Plate XIX, Fig. 9). Tergite and sternite of the 8th somite separated along the pleural margins so that the sternum forms a ventral hood beneath the genitalia; harpes spoon-shaped or truncate, often unsymmetrical and with irregular processes; aedoeagus short; uncus weakly developed, absent or replaced by socii; gnathos usually absent. Lymnaecia, Walshia, Chrysopeleia, Pyroderces and Cosmopteryx agree with these characters.

Gelechiidæ—(Plate XIX, Fig. 7). Eighth sternite and tergite as in the Cosmopterygidæ, the sternite often more elongate and hood-like; entire genitalia usually elongate; aedoeagus long; harpes spoon-shaped or finger-like, usually divided and often unsymmetrical; gnathos and uncus usually present.

Scythrididæ—(Plate XIX, Fig. 10). Much as in the Gelechiidæ, but with a marked tendency toward a reduction of the tegumen and a fusion of it with the aedoeagus, so that the harpes are often the only articulated members in the genitalia.

Conclusions: On the basis of the characters offered by the male genitalia, the Microlepidoptera may be divided into two main groups or suborders comparable to the Jugatae and Frenatae of Comstock and the Homoneura and Heteroneura of Tillyard. The superfamilies Hepialoidea and Micropterygoidea of the Homoneura are readily separable and the families comprising them may likewise be distinguished from each other by characters of the male genitalia. The characters of the greatest importance in the genitalia of the Homoneura are found in the penis and its armature i. e. the aedoeagus and the juxta. Previous investigations by the author (1924) have shown these characters to be of the greatest fundamental importance in that a chitinized tube-like aedoeagus and a well defined juxta are absent or rudimentary in such orders as the Neuroptera and Mecoptera, begin their development in the primitive Trichoptera and Lepidoptera, and reach a high stage of perfection in the higher families of both of these orders.

The Tineoidea of the Heteroneura may be divided into two groups comparable to the Aculeatae and Nonaculeatea of Spuler and the families in each of these divisions possess distinctive characters which vary in importance with the phylogenetic value of the family concerned. The development of a hinged or articulate uncus and gnathos and the reduction in the size of the vinculum are two tendencies most noticeable and of considerable significance in the families of the higher Tineoidea.

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EXPLANATION OF PLATES.

PLATE XVIII.

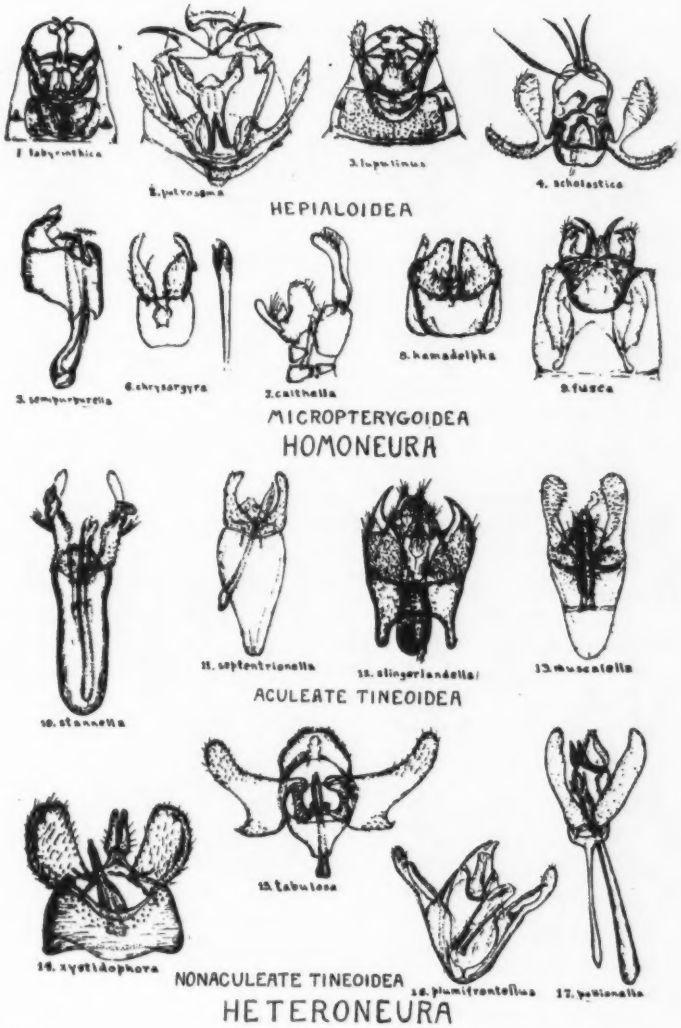
Hepialoidea, Micropterygoidea and Lower Tineoidea.

1. Trictena labyrinthica Don.
2. Prototheora petrosema Meyr.
3. Hepialus lupulinus L.
4. Paleoses scholastica Turn.
5. Eriocrania semipurpurella Stph.
6. Sabatinca chrysargyra Meyr.
7. Micropteryx calthella L.
8. Mnesarchaea hamadelpha Meyr.
9. Mnesarchaea fusca Philp.
10. Heliozela stannella F. R.
11. Adela septentrionella Wlsm.
12. Nepticula slingerlandella Kearf.
13. Incurvaria muscalella F.
14. Tinea xystidophora Meyr.
15. Talaeporia tabulosa Ritz.
16. Acrolophus plumifrontellus Clem.
17. Tinea pellionella L.

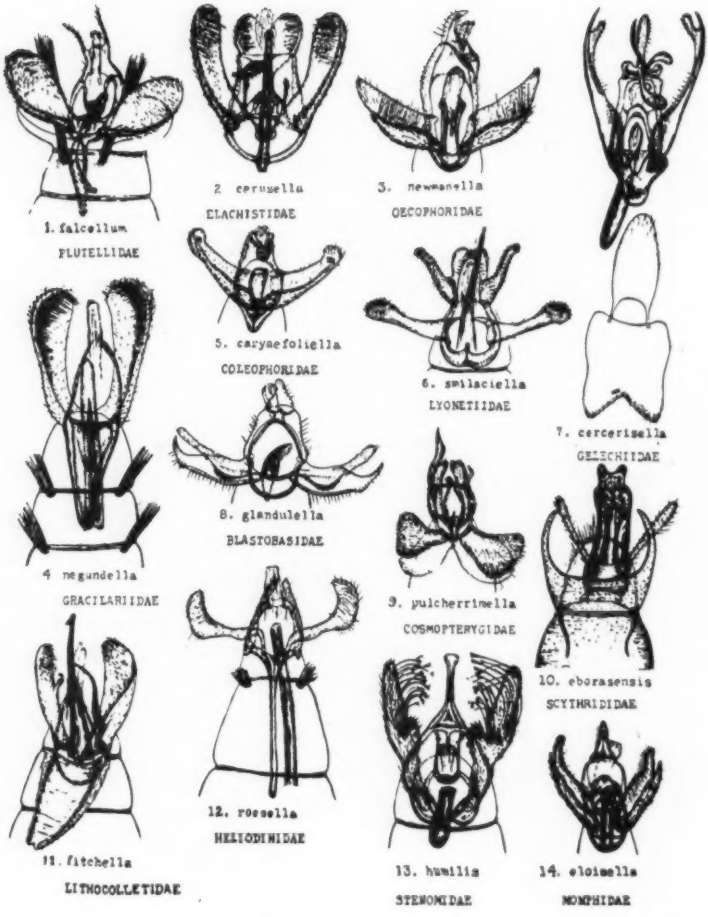
PLATE XIX.

Higher Tineoidea.

1. Cerostoma falcellum Hb.
2. Elachista cerusella Hb.
3. Oecophora newmanella Clem.
4. Gracilaria negundella Chamb.
5. Coleophora caryaefoliella Clem.
6. Proleucoptera smilaciella Bsk.
7. Gelechia cercerisella Chamb.
8. Valentinia glandulella Riley.
9. Cosmopteryx pulcherrimella Chamb.
10. Scythris eborasensis Zell.
11. Lithocolletis fitchella Clem.
12. Heliodines roesella L.
13. Stenoma humilis Zell.
14. Mompha eloisella Clem.


1. labyrinthica
2. petrosema
3. lupulinus
4. scholastica
HEPIALOIDEA
5. semipurpurella
6. chrysargyra
7. caithella
8. hamadelpha
9. fusca
MICROPTERYGOIDEA
HOMONEURA
10. stannella
11. septentrionella
12. clingerlandella
13. muscatella
ACULEATE TINEOIDEA
14. cystidophora
15. tabulosa
16. plumifrontellus
17. pavonella
NONACULEATE TINEOIDEA
HETERONEURA

J. R. Eyer


1. falcellum
FLUTELLIDAE
2 cerusella
ELACHISTIDAE
3. newmanella
OECOPHORIDAE
5. carynefoliella
COLEOPHORIDAE
6. smilaciella
LYONETIIDAE
7. cercerisella
GELECHIIDAE
4 negundella
GRACILARIIDAE
8. glandulella
BLASTOBASIDAE
9. pulcherrimella
COSMOPTERYGIDAE
10. eborasensis
SCYTHRIDIDAE
11. fitchella
LITHOCOLLETIDAE
12. roseella
HELIODINIDAE
13. humilis
STENOMIDAE
14. eloisella
MOMPHIDAE

J. R. Eyer

NOTES ON CUBAN REDUVIIDÆ.

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Santiago de las Vegas, Cuba,

and

S. B. FRACKER,
Madison, Wisconsin.

In a recent paper on neotropical Reduviidae* the authors included all of the Cuban species of this family of which material was then available. Five additional forms have since come to hand from that country; among them two new species, one of which represents a new genus.

Saica rubripes Champion. This Central American species has recently been recorded from Cuba (Cayamas) by McAtee and Malloch.† We have examined two specimens from Havana Province (F. Cervera, collector.) These agree with Champion's figure and description except that they are smaller (12.5 mm.) and the pronotal and scutellar spines are not flavescent but red.

Alloeocranum biannulipes Montr. et Sign. We have four specimens taken at Camaguey (J. Acuna, collector.)

Triatoma rubrofasciata De Geer. This widely distributed species is here included on the basis of a single specimen taken near Havana (F. Cervera, collector.)

Bolbodera new genus.

A specimen in the Gundlach Museum, Havana, labeled "*Bolbodera scabrosa* Uhler" represents a new genus and species as a description of the insect was evidently never published.

Generic Characters: Head large, elongate, moderately broad and strongly produced before eyes, swollen behind; antennae inserted on the lateral or dorso-lateral margins, somewhat nearer apex than eyes, antenniferous tubercles large, projecting strongly from sides of head;

*Notes on Some Neotropical Reduviidæ, by S. B. Fracker and S. C. Bruner; Annals Ent. Soc. Am., 17 (1924), 163–174.

†Notes on American Bactrodinæ and Saicinæ; Annals Ent. Soc. Am., 16 (1923), 247–254.

juga projecting to form a distinct porrect tooth-like process on either side; antennae with first segment short, scarcely attaining apex of head, second segment much longer, third and fourth shorter than second; eyes prominent; ocelli widely separated, placed well behind caudal margin of eyes, not or very slightly elevated. Body rather broad, glabrous; the connexivum considerably expanded. Pronotum with distinct constriction cephalad of center, the anterior lateral angles greatly produced forming a large sub-porrect somewhat flattened tooth at either side of neck; the posterior lateral angles obtuse; the anterior lobe more or less tuberculate, two broad obtuse tubercles or elevations on disc; posterior lobe unarmed. Scutellum with apex narrow, considerably produced. Elytra broad, apex rounded, venation as in Triatoma. Femora somewhat incrassate, armed beneath with several teeth or stout spines; tibiae and tarsi normal.

This genus falls clearly within the subfamily Reduviinae as the pronotum is constricted in front of the middle, the wings are without a quadrangular areole, and the scutellum is normal. The insertion of the antennae and the location of the ocelli show the relation to Triotoma and its relatives. There is a resemblance to Belminus Stal in the thickened and armed femora, but the greatly produced anterior angles of the pronotum and the fact that the scutellum is unarmed at the base are distinguishing points. Meccus shows some similarity in the deeply constricted pronotum and the presence of the tubercles on the anterior lobe. The short antennae and the shape of the apex of the head are, however, characteristic. The size is much smaller than in other relatives of Triatoma.

Bolbodera scabrosa new species. Color flavo-testaceous, black, and red or pink; form broad. Head black, first and second segments of antennae dull brown, third and fourth segments testaceous, infuscated at base; pronotum above flavo-testaceous with base before apex of scutellum infuscated; connexivum red or deep pink, the third and fourth exposed segments with black patch at anterior angle, the remaining posterior segments entirely black; legs black, tarsi flavescent.

Body glabrous, a few minute prostrate hairs on antennae and tibiae towards apex. Head elongate, subequal in length to pronotum, finely tuberculate or granulate, comparatively broad in front of eyes, apex with a porrect tooth (jugum) or stout spine on either side; antenniferous tubercles large, conical, projecting prominently from sides of head; postocular part rather short, considerably swollen, constricted behind, neck smooth; first segment of antennae short, rather stout, scarcely attaining apex of head, second segment about twice as long as first, third segment shorter than second, fourth segment very short; eyes very prominent, of moderate size, ocelli scarcely as widely separated

as eyes, very slightly elevated. Pronotum strongly constricted laterally and above before center; anterior lobe finely tuberculate with two large very obtuse tubercles or elevations on disc, partially divided above by shallow sulcus extending forward from transverse constriction, obsolete before anterior margin; anterior lateral angles produced in front as a large somewhat flattened sub-porrect tooth on either side of neck, about one-half as long as lobe; posterior lobe rather thinly

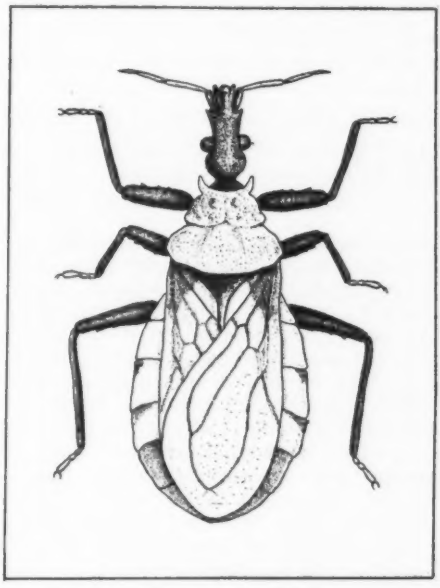

Fig. 1. *Bolbodera scabrosa* new species.

granulate, two low obtuse dorsal carinae, obsolete before posterior margin, lateral angles obtuse, rather prominent, slightly elevated. Scutellum with surface somewhat sculptured and coarsely tuberculate, apex produced as a long obtuse spine. Hemelytra broad, venation strongly marked, base and costal margin roughened. Connexivum broad, rounded. Femora moderately incrassate, very finely and thinly granulate, armed below with several conical teeth. Length, 8–9 mm. (approximate.)

Holotype in the Gundlach Museum.

Zirta luacesi new species. Two specimens from Camaguey represent a new species of Ectrichodiinae. They are more closely related to *Zirta hirticornis* than to other species of the subfamily and are being placed in that genus, which has not heretofore been recorded from the West Indies. The four-segmented antennae separate the new form from the genera Mindarus, Rhiginia (Ectrichodia), and Ectrichodiella. The second antennal segment is about twice as long as the first, as in Pothea, but the short first rostral segment, and the shape of the postocular portion of the head show a closer affinity with Zirta.

The new species does not resemble *Z. hirticornis* in color, as the latter is bright red, with the hemelytra, legs, and most of the abdomen black, while *Z. luacesi* is mottled yellow and brown.

Male.—Shining fusco-piceous and flavescent. Head flavescent above, below fuscous, rostrum flavescent, antennae light fuscous. Anterior lobe of pronotum fusco-piceous, disc with large rounded flavescent macula extending caudad to include impression near posterior border; posterior lobe fusco-piceous, lateral angles pale flavescent, this extending forward and inward as narrow band to transverse suture. Scutellum fuscous. Hemelytra dull fuscous, the basal half flavescent marked with fuscous area extending from anal margin to disc of corium and a smaller triangular patch on disc; veins of membrane pale towards base. Connexival segments each flavescent anteriorly with the posterior half fusco-piceous; anal segment flavescent. Below fusco-piceous; area around insertion of legs and lateral margins of venter flavescent; genital region fuscous. Legs flavescent marked with fuscous as follows: A broad band in center or beyond center of femora, apical and basal thirds of anterior and intermediate tibiae, base and apex of posterior tibiae; the third segment of tarsi lightly infuscated.

Body moderately elongate, glabrous. Head rather broad, straight, considerably longer before eyes, subequal in length to pronotum; postocular portion short, rapidly narrowed posteriorly but not as suddenly constricted as in *Z. hirticornis*. Eyes moderately large, prominent, ocelligerous swelling well elevated; antennae of four segments not subdivided, segment one slightly incrassate, one-half length of head and extending well beyond apex, segment two nearly twice as long as first, segments three and four short, together shorter than second, all with long erect pilosity. Rostrum stout, attaining prothoracic coxae; first segment almost as long as second and third combined, but not reaching cephalic margin of eyes. Pronotum smooth, planate above, transverse constriction distinct, shallow; anterior lobe with large rounded impression on disc near posterior margin; posterior lobe about a third broader than anterior, a median sulcus extending caudad from trans-

verse constriction to about center of disc, posterior lateral angles rounded rather prominent, slightly elevated. Scutellum coarsely sculptured, apex with two well separated blunt points. Connexivum rather narrow. Hemelytra extending to apex of abdomen. Legs short, anterior and intermediate femora somewhat incrassate. Anterior and intermediate tibiae curved, apical third dilated and with mesal surface finely pilose; posterior tibiae slender, straight. Posterior tarsi with the second and third segments subequal in length, first much shorter; all tarsi finely pilose. Length, 11 mm., breadth, 3 mm.

Female.—An almost completely apterous specimen having mere pads like a nymph is very similar in plan of coloration to the above but the pale markings are testaceous rather than flavescent, the anterior lobe of the pronotum is entirely fusco-piceous, and the base of the second antennal segment is flavescent. In form it is much stouter, the head larger and thicker, the eyes much smaller, and the antennae more sparsely and finely pilose. The anterior lobe of the pronotum and the abdomen are also broader, and the legs much stouter. Length, 11.5 mm., breadth, abdomen 4 mm.

Holotype: male, taken at Camaguey, Cuba, August 11, 1921, by J. Acuna, in collection of S. B. Fracker.

Allotype: female, same locality and collector, taken July 15, 1924, in collection of S. B. Fracker.

We dedicate this species to the memory of the late Don Roberto Luaces of Camaguey.

NEW INTERMEDIARY INSECT HOST RECORDS FOR THE GIANT THORN-HEADED WORM OF SWINE.

Phyllophaga rugosa an Intermediary Host, and the Adult Beetle a Carrier.*

ROBERT D. GLASGOW,
University of Illinois.

The giant thorn-headed worm, *Macracanthorhynchus hirudinaceus* (Pallas) Travassos, is a serious, a very common, and a widely distributed intestinal parasite of swine. This parasite is provided with a retractile rostrum, the so-called "head," which is armed with recurved, thorn-like spines or hooks. The worm inhabits the small intestine of the pig, where it anchors itself by working the spiny "head" deeply into the tissues of the intestinal wall. The resulting irritation causes an annular proliferation about the point of attachment, which is marked on the outer surface of the intestine by a firm, somewhat elevated mass of hypertrophied tissue, from six to ten millimeters in diameter. The lesion so produced may develop into an abscess, and may even result in a perforation of the intestinal wall.

The eggs of this parasite are distributed, mixed with the excreta of the infested pig, and are supposed to hatch only when swallowed by certain species of insects. Apparently, also, the worm must undergo the first part of its post-embryonic development in the body of an insect host. When, and presumably only when this so-called larval development of the worm has been completed, a pig may acquire the parasite by eating the infested insect.

In a paper published in 1871, Anton Schneider states that this worm may develop in grubs of the common European May-beetle, *Melolontha vulgaris*, and that the encysted parasite may persist through the transformation of the insect host so that it may occur in the adult of this beetle. It has been

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*Contribution from the Entomological Laboratories of the University of Illinois, Number 95.

(Read at the Kansas City meeting of the Entomological Society of America, December 29, 1925).

shown further, by Kaiser, that in Europe this parasite may develop in the larva of *Cetonia aurata*, and, by Wolffhuegel, that in Argentina it may develop in the larva of *Dilobderus abderus*.

Hitherto, our knowledge of the intermediary host relations of the giant thorn-headed worm in North America has been limited to the work of Dr. C. W. Stiles; who, thirty-odd years ago, showed that this parasite may undergo its larval development in white grubs, which were identified as belonging to the genus *Phyllophaga* (or Lachnosterna), and probably to the species *arcuata* Smith.†

For more than a quarter of a century this interesting problem has been neglected in the United States; its further investigation having been discouraged, doubtless, by the difficulty attending the separation of the *Phyllophaga* species even in the adult stage; by the long life cycle of these insects; and, owing to the lack of any keys based upon the studies of immature stages, by the impossibility of identifying the species in the larva stage. Because of this situation, an interest in medical and veterinary entomology coupled with a knowledge of the May-beetle species‡ has seemed in some measure to place upon the writer an obligation to take up the problem where Stiles left it.

The problem has been approached by the writer from two directions, first through the use of insect larvae reared from eggs deposited by identified females, and second, through rearing experimentally infested larvae of unidentified species to the adult stage for identification. Infestation of the grubs has been readily secured, essentially by the means employed by Stiles; but to permit individual observation and treatment, the grubs have been kept separately, each in a small, tin, ointment box.

Both the following, and additional results of this investigation will be presented in detail in the near future. The present paper is intended only to place on record the fact that the encysted larva of the giant thorn-headed worm has been

†Stiles, C. W. Notes sur les Parasites III. Sur l'hote intermediaire de l'Echinorhynchus gigas en Amerique, Bulletin de la Societe Zoologique de France, 1891, vol. 16, pp. 240-242.

‡Glasgow, R. D., *Phyllophaga* Harris. . . : A Revision of the Synonomy . . . Bulletin of the Illinois State Laboratory of Natural History, 1916, vol. V, pp. 365-379.

found in the adult beetle of *Phyllophaga rugosa* Melsheimer, reared from experimentally infested grubs, and to direct attention to the general significance of that fact. The infested beetles are active, and appear to be perfectly normal in every way. Nothing could be observed to indicate that the encysted parasite might not be transported by the flight of the infested beetles from a contaminated area to a clean field, or to a clean hog lot where this parasite had not previously occurred.

The adult May-beetles are nocturnal in their habits, and upon emerging in the spring fly to, and feed at night upon the leaves of various trees and shrubs. During the day the beetles conceal themselves under the fallen grass in sod covered land, or busy themselves in loose soil; and such places are also visited by the female beetles for oviposition. Here the beetles are sought as diligently by swine as are the grubs, and when found, are devoured as greedily.

The evident probability that larval cysts of the giant thorn-headed worm may be transported from place to place by the flight of the adult May-beetles, introduces a factor in the dissemination of this parasite which should certainly be taken into account in carrying out the so-called McLean county system of swine sanitation. Indeed, dissemination by adults of the intermediary insect host would seem to make the control of this parasite a community problem, rather than a problem for the individual breeder.

HARRY ARTHUR GOSSARD

REPORT OF COMMITTEE TO PREPARE MEMORIAL TO PROF. GOSSARD AND DR. HUNTER.*

Harry Arthur Gossard

Prof. H. A. Gossard was born at Ames, Iowa, Feb. 7, 1868 and died at Wooster, Ohio, Dec. 18, 1925. His boyhood days were spent on a farm near Ames, Iowa, and his early schooling was received in the country schools where he evidenced the possession of a remarkably retentive memory, and thus was able to advance more rapidly than other boys of his own age. At the age of seventeen he was granted a certificate and taught a term of country school near his home. To use Prof. Gossard's own words:—"I always opened school with a Bible reading and the Lord's Prayer and generally followed with some scientific experiment in physics or chemistry to enlist their attention. All the sciences from my earliest childhood had a fascination for me, but I had no scientific books at this time which were of any help to me."

In the spring of 1886 he entered the Iowa Agricultural College and graduated as an honor student in 1889, standing fifth in his class. He also held the distinction of being next to the youngest member of his class. In 1892 the degree of M. S. was granted him by the same institution.

As a graduate student in entomology he was the first man to serve as assistant entomologist in Iowa.

In 1893 he was asked to take the presidency of Albion Seminary at Albion, Iowa. Dr. E. D. Ball was the other member of the faculty for the first year but with the substantial growth in the student body the second year several new faculty members were added.

The natural sciences, however, had a stronger appeal to him than administrative work and from 1895 to 1898 he was

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*This Committee was appointed by President Dean at the Kansas City meeting, and this report was not included in the Proceedings as it was desired to secure portraits.

instructor in Science in the high schools of Rochester, Minnesota, and Princeton, Illinois. At the latter place he met his future wife, Miss Lora H. Clark, who was at the time a fellow teacher and to whom he was married in 1903. Three sons, one who is now a Junior in the Ohio State University and two in high school at Wooster, were born to this union.

In 1898 Prof. Gossard entered into the more serious aspects of his scientific career and particularly as an entomologist, when he was elected to the Professorship of Entomology, Zoology and Geology of the University of Florida, and Entomologist of the Florida Experiment Station. He remained in this position until 1904 when he became Entomologist of the Ohio Agricultural Experiment Station, which position he held until his death. From 1917 he held the title of non-resident Professor of Entomology of the Ohio State University.

As an Entomologist Prof. Gossard was well informed on the subject as a whole and his retentive memory was of inestimable benefit in enabling him to recall without hesitation the technical names of the insects with which he had become familiar and the intricate details of the life histories of economic forms. His contributions to the science were wholly of an economic nature and cover a wide range of subjects as is testified by the many bulletins and articles bearing his name. He wrote easily and readily, with little effort transferring his thoughts in logical sequence to the written page.

At the time of his death Prof. Gossard was President of the American Association of Economic Entomologists. Aside from membership in our Society, Prof. Gossard was a Fellow in the American Association for the Advancement of Science and the Ohio Academy of Science; a life member of the Ohio and Florida Horticultural Societies; a member of the Iowa Academy of Science, the American Nut Growers Association, Sigma Xi and of Psi Kappa Psi. He was Vice President of the Monticello Nurseries Co., of Monticello, Florida, a firm well known in southern nursery circles with valuable nursery holdings and pecan orchards in Florida, Georgia, and Alabama. Because of his financial connections in the south it was his custom to attend the winter meeting of the American Nut Growers Association and his papers with insect problems in southern nurseries and pecan orchards were always received with the greatest interest.

WALTER DAVID HUNTER

Measured from the standpoint of lasting benefit to agriculture the two most outstanding accomplishments of Prof. Gossard's entomological career were his "Ten Years Experiments in Spraying Ohio Apple Orchards" and "The Annual Wheat Field Survey." The publications of the results of the former gave a great impetus to apple growing in Ohio, and the latter not only has resulted in the saving of millions of dollars to Ohio wheat growers but in modified form is now applied in several of the wheat producing states.

In matters of institutional administration Prof. Gossard's judgment was sought and respected; in departmental administration he was patient, kindly and inspiring and in his community he was a Christian gentleman in the truest sense of the word. The sincere sympathy of our Society is tendered to the bereaved family.

Walter David Hunter.

Walter David Hunter, Chief of the Section of Southern Field Crop Insect Investigations of the Bureau of Entomology and member of the Federal Horticultural Board of the Department of Agriculture, died at El Paso, Texas, October 14, 1925. Doctor Hunter was born at Lincoln, Nebraska, December 14, 1875. He was educated at the University of Nebraska, taking his Bachelor degree in 1895 and the degree of Master of Arts in 1897. He was Instructor in that University until 1900, and then for a year was Assistant Entomologist of the Iowa State College of Agriculture. In 1901 he came to the Department of Agriculture at Washington and was sent to Texas to investigate the cotton boll weevil. He speedily built up a strong force of entomologists in Texas, and continued his work with the boll weevil until the time of his death. As his work progressed it was broadened to include, not only all insects affecting cotton, but also those affecting other southern field crops such as sugar cane, rice and tobacco. Later his field also included insects affecting the health of man and animals, and under his direction investigations were begun on a number of important pests of live stock. Investigations of the Rocky Mountain fever tick, of malarial mosquitoes and of the house fly, and (during the war) of body-lice, were carried on.

With the crossing of the Rio Grande by the pink bollworm of cotton, his attention was turned especially towards this insect, and, by virtue of his position on the Federal Horticultural Board, he started an office in Houston, Texas, and devoted his principal energies during the latter part of his life to the prevention of the spread of this destructive pest and to an intensive effort to exterminate it on American soil.

Doctor Hunter's early work while in Nebraska was devoted to a taxonomic study of the Syrphidae and to an annual survey of the conditions relative to the possible swarming of *Melanoplus spretus*.

In economic entomology he achieved a standing of the highest rank. His personal research work was of a very high and sound character, and he carried the research spirit into his administrative work on economic problems. He was a charter member of the Entomological Society of America, but, apparently by some strange oversight, was never made a fellow.

He was President of the Association of Economic Entomologists in 1913, and President of the Entomological Society of Washington in 1914, his retiring addresses before these two societies both relating to medical entomology. He was given the honorary degree of LL. D. by Tulane University in 1913.

A full account of Hunter's life and work will be found in the Proceedings of the Entomological Society of Washington, Volume 27, No. 9 (1925), pages 170-181, and his complete bibliography, covering about one hundred titles and prepared by Miss Mabel Colcord, accompanies it.

He is survived by a widow now living in Houston, Texas.

Doctor Hunter was a notable figure in American entomology, a man of broad knowledge, infinite tact, greatly respected in the South and greatly loved by those who knew him well. All our number will join in sincere sympathy with Mrs. Hunter in her loss.

L. O. Howard,
J. S. Houser,
Herbert Osborn,
Committee.